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The American Journal of Medicine

Vol. XI OCTOBER, 1951 No. 4

Editorial

Clinical Studies

In this important study Dr. Prinzmetal and associates used high-speed cinematographs and direct lead electrocardiograms to investigate, for the first time, the entire course of the excitation wave in auricular flutter as produced experimentally in the dog and as occurring in man. It could be shown that the contraction and excitation waves of auricular flutter arise from the ectopic focus (in man usually in the caudal region of the auricles) and spread outward like ripples in a pond, through the auricles in all available directions simultaneously; no circus movement, as postulated by Lewis, was observed. Moreover, both auricular tachycardia and auricular flutter were found to represent essentially the same disturbance, namely, a discharge of impulses from an ectopic focus at a rate greater than that from the sinus node but lower than the fibrillation threshold; the rate in tachycardia being slower than that in flutter.

This summary of the experience with pronestyl in the treatment of ninety-eight episodes of ectopic rhythm, chiefly ventricular tachycardias and ventricular premature contractions, again indicates the usefulness of this drug in the management of ventricular arrhythmias. The untoward reactions encountered are described in sufficient detail to give added interest to the report.

Isuprel, a useful agent in the treatment of bronchial asthma, was found (like epinephrine) to increase the cardiac output and pulse pressure after subcutaneous administration. The authors therefore advise caution in its use in asthmatic patients with heart disease.

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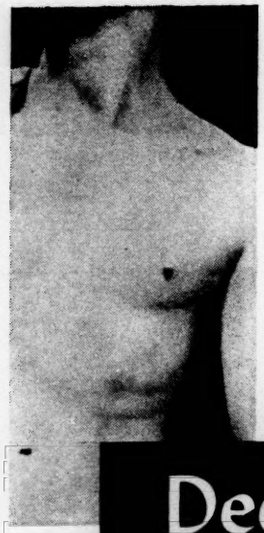

the NEW therapy in "functional G. I. distress"...

Decholin with Belladonna

Patients complaining of gastrointestinal distress without detectable organic cause are common problems in daily practice. By combining spasmolytic action with improvement in liver function, *Decholin/Belladonna* — in such cases — gives symptomatic relief by

reliable spasmolysis

hydrocholeretic flushing of biliary tract

improved blood supply to liver

mild, natural laxation without catharsis

While of special value in functional dyspepsia, *Decholin/Belladonna* is, of course, treatment of choice in biliary tract disorders for thorough and unimpeded flushing of the biliary system.

DOSAGE: One or, if necessary, two *Decholin/Belladonna* tablets three times daily after meals.

PACKAGING: *Decholin* (brand of dehydrocholic acid) *with Belladonna*, bottles of 100 tablets. Each tablet contains dehydrocholic acid 3¾ gr. and belladonna ⅛ gr. (equivalent to tincture of belladonna, 7 minims).

Decholin, trademark reg.

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Diagnostic Criteria of Vascular Headaches

		MIGRAINE	MIGRAINE EQUIVALENTS	MIGRAINE VARIANTS (e.g. Histaminic Cephalgia)	TENSION HEADACHE
BACKGROUND	INCIDENCE	8 to 10 million in U.S.; (ratio of women: men::2.5:1)	As common as migraine	Less common than migraine (mostly in men)	Most common of all headaches
	FAMILIAL HISTORY	Hereditary factor	Hereditary factor	Not significant	Not significant
ONSET OF ATTACK	STAGE OF LIFE	Puberty to menopause	Puberty to menopause	Often after 35	Any
	RATE OF ONSET	Abrupt to gradual	Abrupt to gradual	Abrupt	No pattern
	TIME OF DAY	Usually early morning, diminishing in evening	Variable	Frequent during sleep	Usually at end of day
	DAY OF WEEK, MONTH, ETC.	Most commonly weekends; frequently pre-menstrual and during periods of conflict, tension or stress.	Same as classical migraine	Occur in series, as often as 10 to 15 times a day, often seasonal.	Variable, but usually preceding or following emotional stress or conflict
DESCRIPTIVE QUALITIES OF HEADACHE	DURATION	Minutes to days	Minutes to days	Under 1 hour	No pattern
	NATURE OF HEAD PAIN	Throbbing or pulsating		Constant, boring	Sustained dull or sharp pain
	SITE	Anywhere in head and face—most commonly at right temple, usually unilateral	*SEE NOTE BELOW	Involves eye, neck and often face; unilateral	Most intense in neck, shoulders and occiput—may spread to frontal region—unilateral or bilateral
	TENDERNESS (RESIDUAL)	Near large extra-cranial arteries. Affected region may involve nasal, paranasal, teeth, ear, neck.	Varies according to location	No residual tenderness	Any of head and neck muscles
	EFFECT OF MANUAL PRESSURE	Pressure upon temporal, frontal, post-auricular arteries often reduces intensity	*NOTE Many cases in this classification suffer pain in regions other than the head	Similar to migraine (classical)	Pressure upon tender muscles or regions of tenderness intensifies headache
	EFFECT OF POSITION OF HEAD	Erect position relieves—shaking head aggravates		Pain eased by sitting up and leaning forward	Shaking head reduces intensity by extending muscles
ASSOCIATED SYMPTOMS OF ATTACK	VISUAL DISTURBANCES	Scintillating scotomata; unilateral homonymous hemianopia usually of short duration preceding attack. Photophobia common	Same as classical migraine Photophobia always present in ophthalmic migraine	Photophobia often present	Partial closure of eyes due to muscle spasm. May give impression of faulty vision; Photophobia common
	OTHER OCULAR SYMPTOMS	Injection of conjunctiva and sclera, lacrimation and ptosis of eyelid may occur	Ptosis of eyelid occurs with ophthalmoplegic migraine	Conjunctival injection, lacrimation, nasal stuffiness, discharge	Injection of sclera and conjunctiva; lacrimation may also occur
	VERTIGO AND OTHER SENSORY DISTURBANCES	May be present; paresthesias of hands and face may precede	Paresthesias of hands and face may precede	Infrequent	Infrequent
	MOOD CHANGES	Depression-irritability precede; exaltation follows	Same as classical migraine	No significant changes	Tension, irritability; desire for attention
	GASTRO-INTESTINAL DISTURBANCES	Anorexia, nausea and vomiting at height of attack; flatulence and distension	In gastro-intestinal migraine - flatulence, distension, constipation	Infrequent	Flatulence and distension common especially with severe headache
	CHANGE IN BODY TEMPERATURE	Slight; sometimes unilateral sweating	In some cases up to 104°F.	Not significant	Chills and hot flashes without change in temperature
	TREATMENT	Emotional guidance, general improvement in health; Endocrine therapy. Specific for attack: Cafergot	Emotional guidance, general improvement in health; Specific for attack: Cafergot; vaso-constrictor agent		Sedatives

The information presented in this table has been compiled by the Sandoz Scientific and Research Staffs from the following publications:

AYASH, J. J.: Journal Lancet *69:* 389 (Nov.) 1949. COHEN, S. G., and CRIEP, L. H.: New England J. Med. *241:* 896 (Dec. 8) 1949. FRIEDMAN, A. P.: Am. Pract. *1:* 948 (Sept.) 1950. FRIEDMAN, A. P., and BRENNER, C.: Am. Pract. *2:* 467 (March) 1948. HANSEL, F. K.: Ann. Allergy *6:* 151 (March-April) 1949. HILSINGER, R. L.: Christ Hospital Bulletin *2:* 85 (April) 1949. MACNEAL, P. S.: Philadelphia Medicine *44:* 1869 (July 9); 1905 (July 16) 1949. MOENCH, L. G.: Headache, Chicago, Year Book Publishers, 1948, chap. 8, 9 and 10. SHEA, J. J.: J. Omaha Mid-West Clin. Soc. *11:* 71 (Aug.) 1950. WALSH, F. M.: Journal Lancet *70:* 259 (July) 1950. WOLFF, H. G.: Headache and Other Head Pain, New York, Oxford University Press, 1948.

Sandoz Pharmaceuticals

DIVISION OF SANDOZ CHEMICAL WORKS, INC.
68 CHARLTON STREET, NEW YORK 14, NEW YORK

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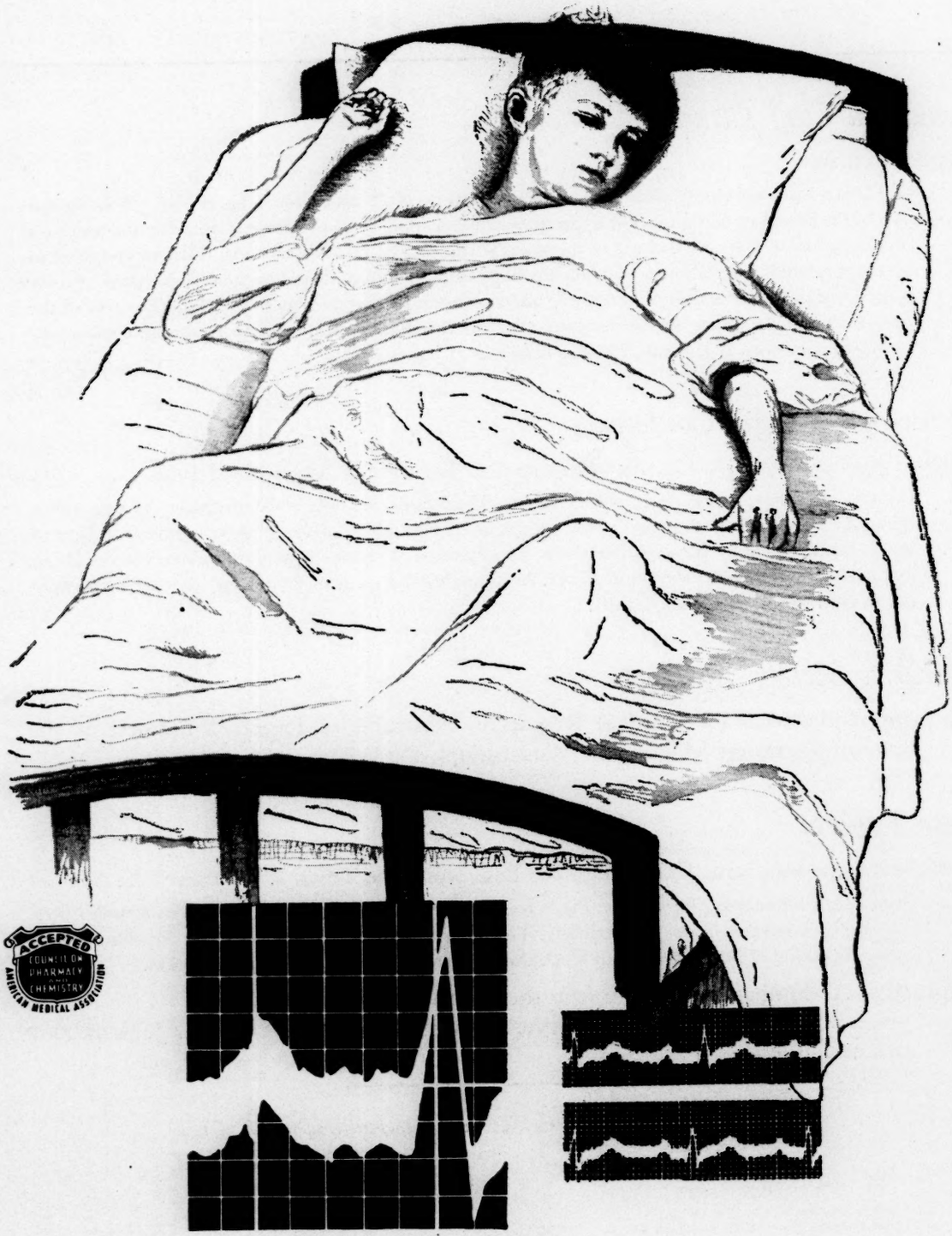
Clinico-pathologic Conference

Research Society Abstracts

Case Reports

Change of address must reach us one month preceding month of issue.

a promising outlook



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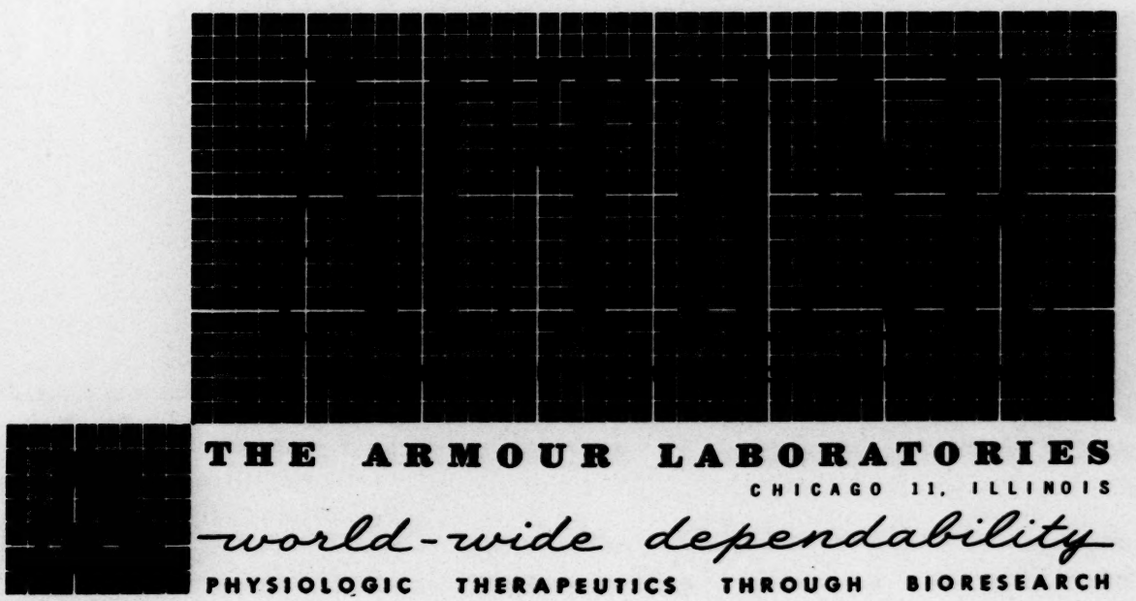
in acute rheumatic fever

ACTHAR, instituted early and in adequate dosage, gives promise of lasting results in rheumatic fever.

ACTHAR, in an increasing number of rheumatic fever patients, has shortened the course of the disease, minimized residual cardiac damage and probably reduced mortality. Systemic signs and symptoms of rheumatic fever usually disappeared within three days—the acute rheumatic process was brought under control, and the electrocardiogram and enlarged heart returned to normal, with regression of pathologic murmurs. Marked cardiac failure, however, necessitates special caution, since sodium and water retention may be produced.

ACTHAR is available in vials of 10, 15, 25 and 40 U.S.P. Provisional Units. One milligram of the Armour Standard for ACTHAR is now accepted as the International Unit; the biologic potency of one International Unit is equal to the biologic potency of one U.S.P. Provisional Unit.



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world-wide dependability

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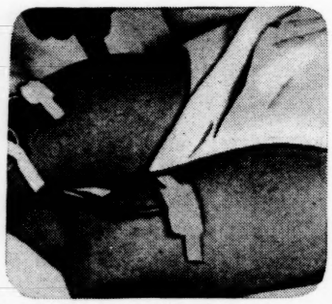
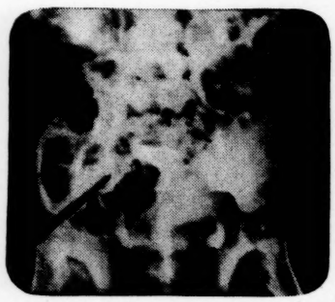
for Medicine . . . Produced with care . . . Designed for health



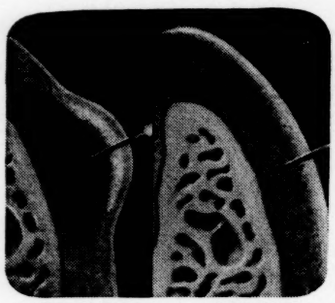
HYPODERMOCLYSIS

more efficient, less painful



CONTRAST MEDIA INJECTED HYPODERMALLY

affords quick, clear visualization



LOCAL ANESTHETIC

rapidly diffuses around nerve area; swelling is minimized

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BECAUSE IT BREAKS

DOWN

THE BARRIER

to diffusion of fluids, WYDASE facilitates hypodermoclysis, local anesthesia, and hypodermal injection of therapeutic agents.

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HYALURONIDASE WYETH

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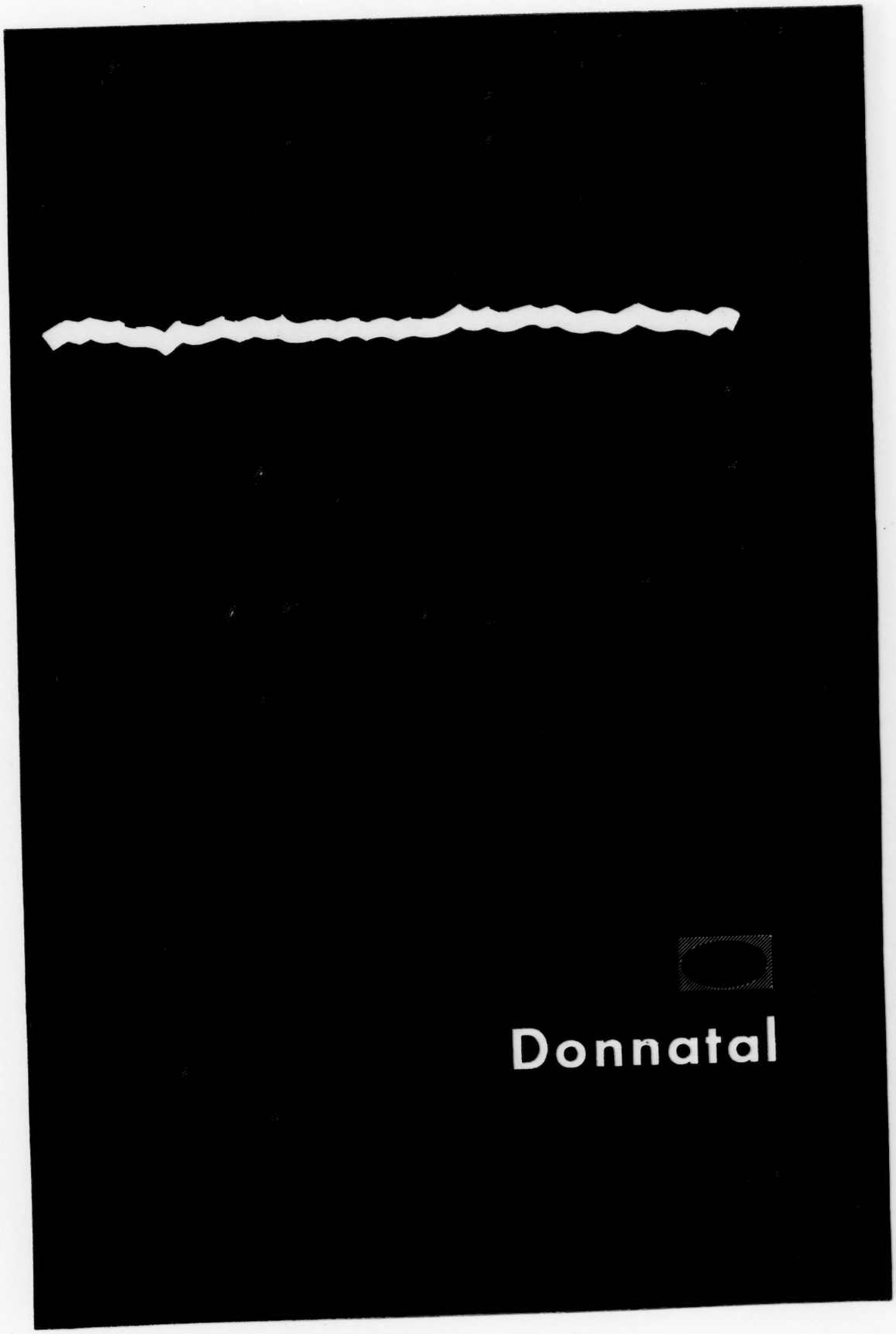
Donnatal

For the Common Anemias...

The Squibb RUBRA Family

	RUBRAMIN *per cc.*	RUBRAFOLIN *per capsule*	RUBRATON *per teaspoonful*	RUBRAFERATE *per capsule*
VITAMIN B_{12}	15, 30 & 50 micrograms	25 micrograms	4.17 micrograms	4.17 micrograms
FOLIC ACID		1.67 milligrams	0.28 milligrams	0.28 milligrams
IRON			220 milligrams ferric ammonium citrate	130 milligrams ferrous sulfate exsic.
VITAMIN C				50 milligrams
DOSAGE THERAPEUTIC	15 to 30 micrograms daily for a week or more; when neurologic involvement is present, 50 micrograms or more daily.	1 or 2 capsules daily	2 teaspoonfuls t.i.d.	2 capsules t.i.d.
MAINTENANCE	Generally, 30-to 50 micrograms twice a month; when neurologic involvement is present, 50 micrograms a week.	1 capsule daily	1 teaspoonful t.i.d.	1 capsule t.i.d.
SUPPLY	1 cc. ampuls, 15 & 30 micrograms of vitamin B_{12} per ampul. 5 & 10 cc. vials, 30 micrograms per cc. 10 cc. vials 50 micrograms per cc.	Bottles of 100	Pint and gallon bottles	Bottles of 100

NOTE: The above are average doses. As with all antianemia preparations, dosages must be adjusted to meet the needs of the individual patient.

Also available: Solution Rubramin Crystalline (Squibb Crystalline Vitamin B_{12} Solution) in 1 cc. ampuls, 15 micrograms of crystalline vitamin B_{12} per ampul, and 10 cc. vials, 30 micrograms of crystalline vitamin B_{12} per cc.

SQUIBB MANUFACTURING CHEMISTS TO THE MEDICAL PROFESSION SINCE 1858

No activity pause at her menopause

Your patient may continue her normal activities even to the extent of keeping pace with her daughter. She will be greatly encouraged, especially when the effectiveness of therapy measures up to expectations. In estrogen therapy an especially useful product . . . . . is:

BENZESTROL

2,4 (p-hydroxyphenyl) –3–ethyl hexane

"Liver function tests, blood studies and urine examinations showed no toxic effects of the synthetic substance BENZESTROL"*

Supplied:

Oral: *Benzestrol* Tablets
0.5 Mg., 1.0 Mg., 100's & 1000's, 2 Mg., 5 Mg. — 50's — 100's — 1000's.

***Benzestrol* Elixir:**
15 Mg. per fluid ounce, Pint Bottles.

Intramuscular: *Benzestrol* Solution in Oil; Aqueous Suspension with 5% Benzyl Alcohol 5.0 Mg. per cc. 10cc Vials.

Local: *Benzestrol* Vaginal Tablets 0.5 Mg. 100's.

AVERAGE DOSE: Menopause — 2 to 3 Mg. daily orally or ½ to 1cc parenterally every 5 days.

Professional Samples and Literature upon Request

NOTE:
Frequently, medication other than estrogens may be required during the menopause. Pleasant tasting Elixir Benzestrol is compatible with many substances.

*Reference: MacBryde, C. M., et. al., A New Synthetic Estrogen, J.A.M.A., 123: 261: 264-(10-2) 43.

Schieffelin & Co. **20 Cooper Square, New York 3, N. Y.**

triple sulfonamides + penicillin

HERE—in an attractive, sweetened, cube-shaped tablet—is a new, highly effective antibacterial combination: 0.1 Gm. each of sulfadiazine, sulfamerazine and sulfamethazine, bolstered with 50,000 units of penicillin.

TRUOZINE WITH PENICILLIN *Dulcet* tablets are as easy to take as candy. Yet clinical evidence indicates that combined sulfonamide-penicillin therapy produces alternating blood level peaks, reducing the danger of development of bacterial resistance to any of the agents administered.[1]

Now, in this season of bacterial infections, give your small patients the benefit of this potent, pleasant therapy. TRUOZINE WITH PENICILLIN *Dulcet* tablets are available in bottles of 25. *To meet increased adult requirements*, prescribe unflavored, grooved TRUOZINE WITH PENICILLIN tablets; each contains 0.5 Gm. of combined sulfadiazine, sulfamerazine and sulfamethazine (equal parts), plus 150,000 units of penicillin. In bottles of 25 and 100.

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with **PENICILLIN**

50,000 units

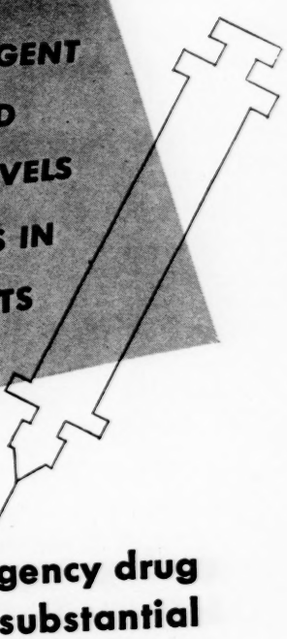
Dulcet TABLETS

1. Vollmer, H., Pomerance, H., and Brandt, K. (1950), Treatment of Pneumonia with a Single Oral Dose of a Penicillin-Sulfadiazine-Sulfamerazine Combination, N. Y. State J. Med., 50:2293, Oct. 1.

A New Achievement in the Control of Hypertensive Crises

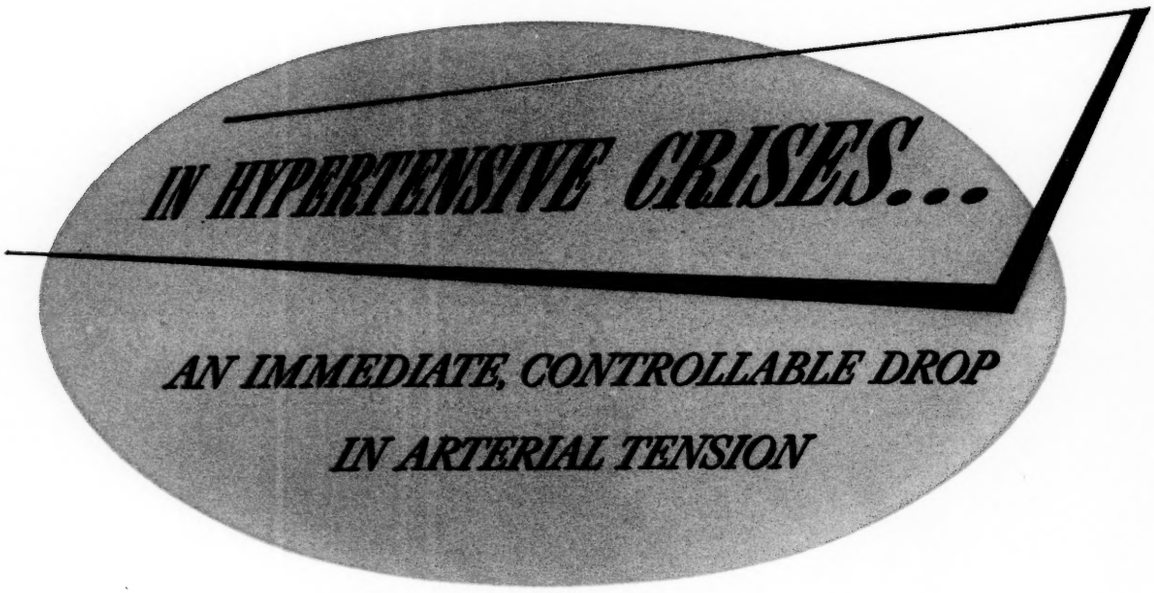
*Veriloid** INTRAVENOUS

A SAFE, POWERFUL HYPOTENSIVE AGENT CAPABLE OF DROPPING THE BLOOD PRESSURE TO NORMOTENSIVE LEVELS WITHIN A MATTER OF MINUTES IN A VAST MAJORITY OF PATIENTS



An invaluable emergency drug for immediate and substantial reduction of the arterial tension

*Trademark of Riker Laboratories, Inc.



VERILOID INTRAVENOUS

Veriloid Intravenous—a biologically standardized hypotensive fraction of Veratrum viride—is a new, highly potent vasorelaxing agent. Infused in proper dosage, it is capable of lowering the arterial tension through central action in a large majority of patients, regardless of the severity or nature of the hypertension. Its action is discernible in a matter of minutes, hence it is an invaluable emergency drug. After the desired blood pressure has been produced, the effect of Veriloid Intravenous can be maintained for hours or even days by intravenous infusion.

Since Veriloid Intravenous makes possible immediate reduction of both systolic and diastolic levels to normal or near-normal limits, it is indicated in the emergency treatment of hypertensive states accompanying cerebral vascular disease, malignant hypertension, hypertensive crises (encephalopathy), and hypertensive states following coronary occlusion.

Veriloid Intravenous is supplied in 5 cc. and 20 cc. ampuls, each cc. containing 0.4 mg. of Veriloid standard reference powder. Complete information regarding dosage and administration is contained in the circular which accompanies each ampul of Veriloid Intravenous. Additional literature will be promptly supplied on request.

RIKER LABORATORIES, INC., 8480 BEVERLY BLVD., LOS ANGELES 48, CALIF.

BLOOD PRESSURE REDUCED

Hypertensive patients benefited 3 ways

STOLIC® and STOLIC® FORTE Tablets bring about a decrease in systolic pressure of approximately 35 mm. of mercury. Pressure begins to fall 15 to 30 minutes after administration; maximum reduction in pressure is produced in about 2½ to 3 hours. The effect of a dose lasts 4 to 6 hours.

Because of their prolonged action, STOLIC Tablets make it possible to maintain steady effects without the need for frequently repeated doses. This is an important advantage in the management of patients with essential hypertension and other circulatory disturbances in which it is desirable to lower the general blood pressure over an extended period of time.

STOLIC Tablets benefit hypertensive patients in 3 ways:

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MANNITOL HEXANITRATE

Mannitol hexanitrate has a prolonged vasodilating action by virtue of its relaxing effect on the smooth muscle of the arteries. This action results in a gradual decrease in arterial pressure; diminished pressure is seen for 4 to 6 hours after administration.

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Delvinal alleviates anxiety and tension, makes patients less apprehensive and less irritable. This tends to prevent fluctuations in blood pressure of emotional origin. Delvinal also contributes to the control of vasomotor function by subduing nerve-reflex excitability.

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RUTIN

Rutin has been found useful in the treatment of increased capillary fragility associated with hypertension. Since cerebral and retinal hemor-

Fall in blood pressure begins 15 to 30 minutes after administration of Stolic. Effect of dose lasts 4 to 6 hours.

rhages occur more frequently when hypertension is accompanied by increased capillary fragility, the administration of rutin is a logical procedure for guarding against such vascular accidents.

COMPOSITION

STOLIC® TABLETS	*Each* STOLIC *Tablet contains:*	
	Mannitol hexanitrate	15 mg. (¼ grain)
	Rutin	20 mg. (⅓ grain)
	Delvinal vinbarbital	30 mg. (½ grain)
STOLIC Forte® TABLETS	*Each* STOLIC FORTE *Tablet contains:*	
	Mannitol hexanitrate	30 mg. (½ grain)
	Rutin	20 mg. (⅓ grain)
	Delvinal vinbarbital	30 mg. (½ grain)

Dosage

The recommended dose for adults is 1 to 2 tablets at intervals of four to six hours. If the systolic pressure is excessively elevated, the dose may be increased in accordance with clinical judgment.

Packaging

STOLIC and STOLIC FORTE Tablets are supplied in bottles of 100 and 1,000.

SHARP & DOHME, Philadelphia 1, Pa.

Verified by Clinical Evidence—
Safer, More Dependable Cardiac Therapy

Myocardone

Announced at the 41st annual meeting of the American Society for Pharmacology and Experimental Therapeutics, MYOCARDONE marks another important advancement in the search for the hormonal control of cardiac function.

Carefully controlled clinical investigations suggest that MYOCARDONE—a new derivative of heart muscle—notably improves the functional efficiency of the heart through cardiotonic and vasodilator actions.

The most recent study involved a group of 58 patients with various cardiac disturbances including:

Cardiac Decompensation — 24 patients with dyspnea, dependent edema, pulmonary congestion, orthopnea and hepatomegaly.

Results with MYOCARDONE: More than half showed definite improvement.

Hypertension Without Decompensation —10 patients with symptoms due to hypertension (dizziness, headache, throbbing, visual disturbance).

Results with MYOCARDONE: ". . . 6 of the 10 enjoyed relief of the distressing symptoms when taking MYOCARDONE . . ."

Compensated Arteriosclerotic Heart Disease — 5 patients who had compensated on routine management.

Results with MYOCARDONE: "Three (60%) of the compensated arteriosclerotics maintained compensation with MYOCARDONE alone."

Angina Pectoris—19 patients with precordial pain aggravated by exertion and emotional tension.

Results with MYOCARDONE: "Definite improvement resulted in two-thirds of the anginal cases."

This report further emphasized:

Efficacy ". . . When MYOCARDONE was administered there was definite subjective and clinical improvement."

Safety "There were virtually no side effects." "All patients tolerated MYOCARDONE well."

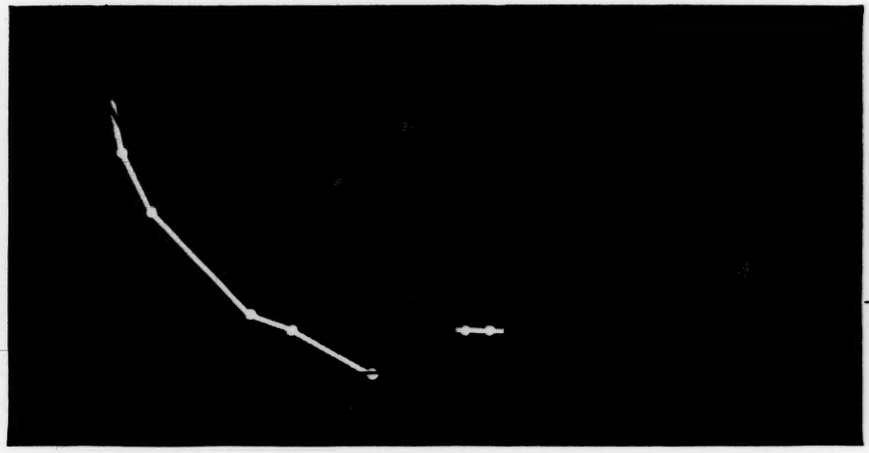
Prolonged Results "Patients whose response to MYOCARDONE was satisfactory continued to do well for from 2 weeks to several months after the drug was withheld."

Digitalis-Nitrite Replacement MYOCARDONE advantageously replaces or supplements digitalis therapy. Reduces or eliminates the need for nitrites in angina pectoris.

Literature on request

Chemico **LABORATORIES, INC.**
Indianapolis 20, Indiana

"true synergism" in an antibiotic combination



Potentiation of penicillin action by addition of streptomycin: in vitro effect on enterococci

Adapted from Jawetz, E.; Gunnison, J. B., and Coleman, V. R.: Science 111:254 (March 10) 1950.

***in vitro*:** "The combined effect of streptomycin and penicillin on enterococci is evidently more than a summation of the individual drug effects... the increased effect... must be a true synergism of the two drugs."[1]

***in therapy*:** "From the results obtained [in 8 cases of subacute enterococcic endocarditis] there can be little doubt that the concurrent administration of penicillin and streptomycin provides highly effective therapy. Moreover the results are obtained without... using massive doses of penicillin..."[2]

In each single dose vial

Combiotic* P-S

Penicillin and dihydroStreptomycin

provides for intramuscular injection, the synergistic action of:

crystalline procaine penicillin G ..... 300,000 units
buffered crystalline sodium penicillin G ... 100,000 units
dihydrostreptomycin (as the sulfate) ..... 1 Gm.
for one 3 cc. aqueous injection, easily prepared by the addition of sterile aqueous diluent.

Combiotic P-S is supplied in a special silicone treated "drain-clear" vial.

1. Jawetz, E.; Gunnison, J. B., and Coleman, V. R.: Science *111*:254 (March 10) 1950.
2. Tompsett, R., and McDermott, W.: Am. J. Med. 7:371 (Sept.) 1949.

*Trademark

ANTIBIOTIC DIVISION **Pfizer** CHAS. PFIZER & CO., INC., *Brooklyn 6, N. Y.*

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Scored 0.5 Gm. tablets.
Bottles of 100 and 1000.

- Remarkably low incidence of side effects—less than 5%
- Lowest acetylation yet reported—less than 10% in blood
- New improved solubility
- Renal complications rare—alkalis not needed
- High, sustained blood levels

WIDE ANTIBACTERIAL SPECTRUM

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A NEW TREATMENT* FOR

Chicken pox

(VARICELLA)

A clinical study of fourteen cases of chicken pox indicated that pruritis, fever and discomfort were promptly relieved with Protamide. No other supportive therapy was used. Protamide was administered by intramuscular injection (½ ampul—0.6 c.c. to one ampul—1.3 c.c.) to all patients daily for two days. The youngest child was 5 months and the oldest 12 years. In all cases to quote—"there was prompt relief of pruritis, fever, discomfort and definite amelioration and shortening of the disease."

Protamide is a sterile colloidal solution of processed and denatured proteolytic enzyme and the dramatic results obtained in the treatment of herpes zoster suggested its value in the treatment of chicken pox.

Reprints covering clinical data on Protamide therapy for chicken pox, herpes zoster and the lightning pains and ataxia of tabes dorsalis are now available to physicians.

**A reprint from the Ohio State Medical Journal, Vol. 47, No. 1 January, 1951, "A New Treatment for Chicken Pox and Other Virus Diseases" by Henry W. Lehrer, M. D., David R. Lehrer, M. D. and Henry G. Lehrer, M. D. will be supplied on request.*

PROTAMIDE

Also Clinically Proved for HERPES ZOSTER and the Lightning Pains and Ataxia of TABES DORSALIS

SHERMAN LABORATORIES
C. H. Sherman, M. D., Founder
BIOLOGICALS • PHARMACEUTICALS
Detroit 15, Michigan

NOT A SALT SUBSTITUTE

For edema control.

Sodium withdrawal—
without sodium depletion

NATRINIL
POWDER

No offensive odor or taste.
Maximum palatability.
Minimum dosage.
Great exchange capacity.
Fine texture.
Mixes readily.
Less bulk required.

Natrinil prevents edema formation by withdrawing sodium from the gastrointestinal tract. Natrinil is indicated in the management of congestive heart failure, hypertension, cirrhosis, or whenever a "salt-free" or a low sodium diet is required. Natrinil allows a more normal diet.

A Cation Exchange Resin of the Carboxylic Type
Hydrogen Cycle 80%
Potassium Cycle 20%

Natrinil Powder
Available, bottles of 10 oz.
Individual packets of 10 Gm. each,
boxes of 24.

THE NATIONAL DRUG COMPANY
Philadelphia 44, Pa.

More Than Half A Century of Service to the Medical Profession

Problem Patient

***quadragenarian** cut-up . . .*

CUNNINGHAM CUDDLE rollicks and romps to homecoming rallies and class reunions, resolved to rekindle the embers of youth with a wassail or two for old Siwash.

Meanwhile, Mr. Cuddle belabors the quality of the whiskey these days and carps about cardialgia.

Not that good old Cuddle, '25, could be considered a clinical curiosity in this age of chronic escapists. On the contrary, he merely typifies another common species of familiar problem patients — the superannuated sophomores whose soma aren't quite up to their psyche. Like the hurriers and worriers and bridge-luncheon butterflies, they bedevil their doctors with a sad lament anent chronic "indigestion," but cling to their merry or morbid course defying the dues of indulgence.

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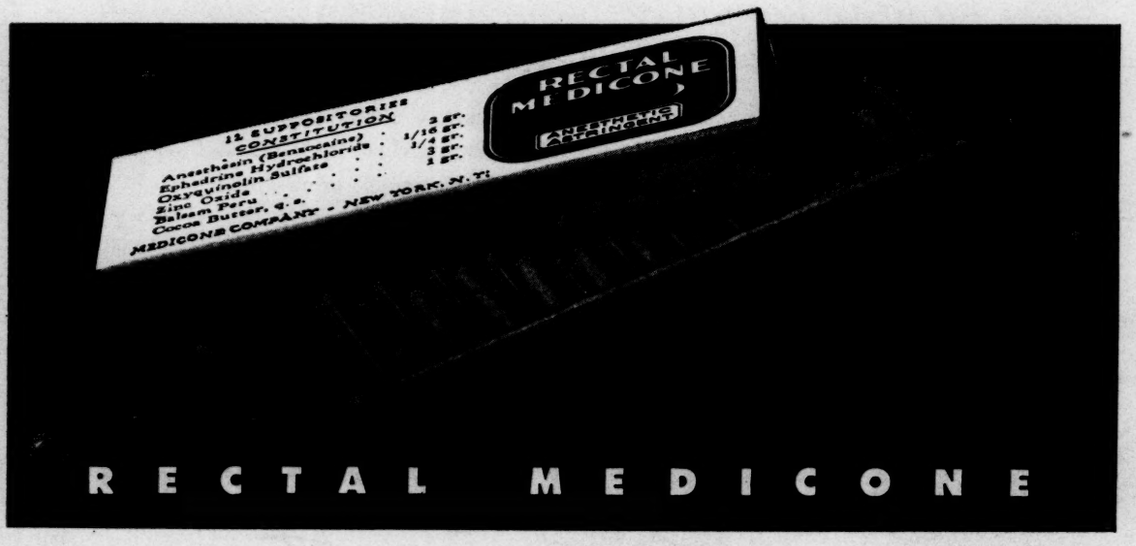

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1. McClintock, J. C.: N. Y. State J. Med. 1951 (In Press)

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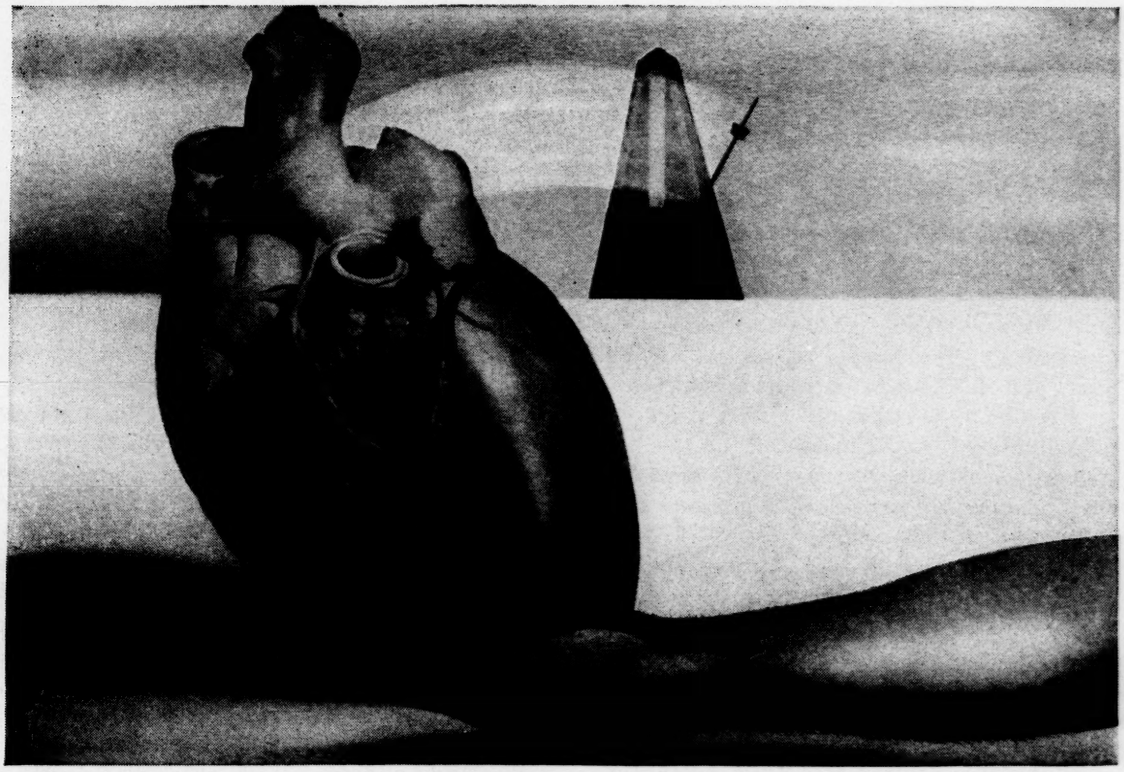

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The American Journal of Medicine

VOL. XI OCTOBER, 1951 No. 4

Editorial

Convalescent Care

CONVALESCENCE, the period of recovery following acute illness or injury, begins early after the acute phase of illness and continues until complete cure or rehabilitation has been attained. The likelihood of important improvement differentiates it from long term illness in which recovery is retarded or remote.

Although recovery actually begins quite early in illness, convalescence is usually considered to commence, quite arbitrarily, immediately after the discharge of the patient from the hospital. Most patients still require medical treatment after leaving the hospital but they are often left to shift for themselves and highly technical and expensive medical care may thus be nullified. Moreover, the conflicts and tensions produced by the sudden transition from a completely protected hospital patient to that of a bewildered, unsheltered discharged person who, in addition to the residue of illness, often is forced to cope with pressing economic and social problems, are potential sources of psychosomatic disorders sometimes more serious than the original illness. Those responsible for the use of public funds for medical care have been concerned for some years about these problems of patients, particularly in the underprivileged group, who have terminated the acute or hospital phase of illness but have not yet achieved the state of well-being, physical and mental, which permits them to function efficiently in their particular social and economic environment.

Despite obvious need, utilization of many of the convalescent facilities which are available has been far below capacity. Most convalescent homes operate as independent units, guarded by rules of admission which seem necessary and equitable to them but which in many instances act as an effective barrier to the admission of those patients who are most in need of their services. This produces a gap in the continuity of complete rehabilitation.

Mounting hospital costs, particularly during the current inflationary period, make early discharge or transfer from hospital to the lower cost convalescent institution desirable if not mandatory. Such recent developments as early ambulation and the use of potent antibiotics have made possible a marked reduction in the necessary in-hospital stay of many patients recovering from serious illness. These factors offer an opportunity to the convalescent institution to complete, at considerably reduced cost, the treatment initiated by the hospital.

The hospital which discharges the patient early, as well as the home which accepts this patient for convalescent care, both share the responsibility for return of the patient to society at the optimum level of usefulness. The activities of both institutions should be closely integrated so that there is continuity of medical care throughout both phases of illness. Good planning through the transition period can change an attitude of apprehension and distrust in the patient to one of realization that transfer to the convalescent home represents a definite milestone of progress along the road to complete recovery.

It is doubtful whether such teamwork can be achieved with maximal efficiency between independent and unrelated medical institutions. Some form of corporate relationship or affiliation is important. Where such an affiliation between hospital and convalescent home exists today, certain definite advantages have been demonstrated for the patient and both of the institutions involved.

Modern concepts of the care of convalescent patients emphasize the need for continuity of medical supervision. Ample experience is now available to demonstrate that this objective is furthered by some form of affiliation between the hospital and the convalescent home.

HERMAN ZAZEELA, M.D.
The Mount Sinai Hospital, New York City
The Neustadter Convalescent Home

Clinical Studies

Auricular Flutter*

MYRON PRINZMETAL, M.D., ELIOT CORDAY, M.D., ROBERT W. OBLATH, M.D.,†
H. E. KRUGER, I. C. BRILL, M.D., JOSHUA FIELDS, M.D., S. REXFORD KENNAMER, M.D.,
JOHN A. OSBORNE, M.D., L. ALLEN SMITH, M.D., ALVIN L. SELLERS, M.D.,
WALTER FLIEG, M.D. *and* EILEEN FINSTON

Los Angeles, California

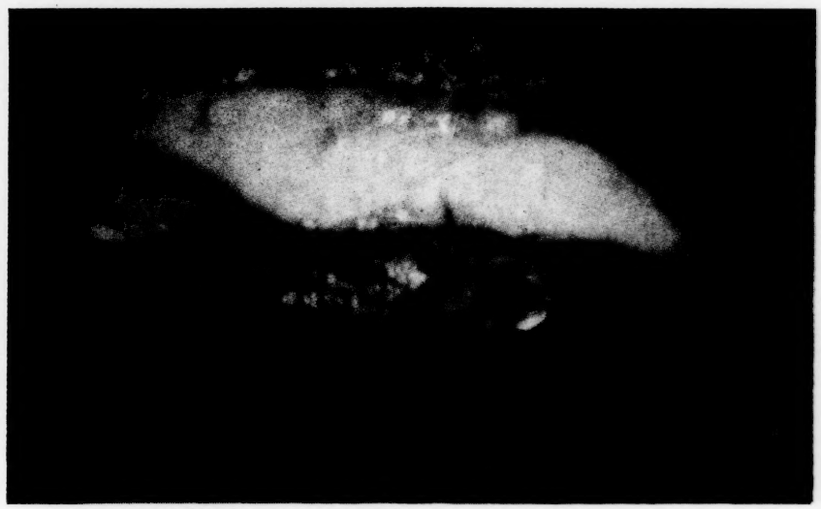
AURICULAR flutter is a relatively rare cardiac disorder characterized by a rapid auricular rate, usually from 260 to 320 per minute, and the almost invariable presence of auriculoventricular block. Despite the low incidence and general benignancy of this arrhythmia its mechanism has been studied extensively by physiologists as well as cardiologists since the turn of the century. MacWilliam[1] in 1887 first used the term "flutter" to describe an experimentally produced disturbance in the dog's auricle; he suggested that the rapid contractions were caused by a series of impulses initiating from a single ectopic focus. Ritchie[2] and Gibson[3,4] in 1905 and Hertz and Goodhart[5] in 1908 described a similar disturbance in the human auricle. Jolly and Ritchie[6] in 1911 first applied the term of auricular flutter to the clinical arrhythmia and established the diagnosis by electrocardiography.

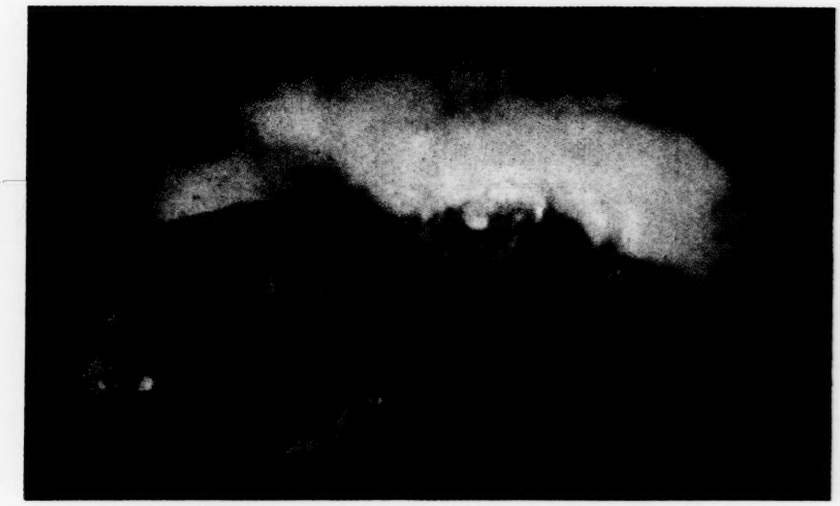
During the past thirty years the prevailing concept of the mechanism of auricular flutter has been the circus movement theory advanced by Sir Thomas Lewis and associates[7–14] in 1918. According to Lewis a main excitation wave is propagated through the fluttering auricles via a narrow circular pathway embracing the two venae cavae; the main wave pursues a unidirectional course and re-enters the original pathway upon completion of each circuit. Small "daughter" waves are presumed to arise from the main wave from whence they spread over the remaining auricular musculature. Although earlier investigations of "circulating rhythm" in isolated muscle provided a basis for Lewis' work the circus movement theory was derived primarily from extensive direct lead electrocardiographic studies of electrically induced flutter in the dog's auricle. Few observations were made in man. Unfortunately, the operative technic employed in Lewis' laboratory failed to permit exposure of the body of the left auricle, thus limiting direct observation to the auricular appendices and right auricular body. The significance of this gap in his determinations was acknowledged by Lewis in the following statement: "No doubt remains as to the course taken by the excitation wave (of flutter) insofar as the exposed surface of the right auricle, the left surface of the superior cava, the band and left appendix are concerned. The weakness obviously lies in the length of the gap between the intra-auricular band and the inferior cava on the return journey."[9]

Several investigations of the mechanism of auricular flutter have been undertaken since Lewis' study. Brams and Katz[15] severed the hypothetical circus pathway in the dog's auricle during electrically produced flutter; the arrhythmia failed to terminate. Weiner and Rosenbleuth[16] and Selfridge[17] noted certain inconsistencies in Lewis' mathematical calculations but nevertheless accepted the circus movement theory as the most probable mechanism of auricular flutter. Scherf[18–20] and associates produced flutter by direct application of aconitine to the auricular wall and demonstrated that cooling or clamping the aconitine focus ter-

* From the Institute for Medical Research, Cedars of Lebanon Hospital, Los Angeles, California, and the Department of Medicine, University of California School of Medicine, Los Angeles, Calif. This study was aided by grants from the L. D. Beaumont Trust Fund, the Blanche May Memorial Fund, the Margaret Mayer Fund, L. Spitz, I. Berlin and E. Mannix. This work was presented at the Los Angeles Post-Graduate Cardiac Meeting, November 3, 1947; the American College of Physicians, April 23, 1948; the American Society for Clinical Investigation, May 2, 1949; the New York Heart Association, February 23, 1949; and the California Medical Association, May, 1949.

† This work was performed during tenure as a Research Fellow of the American Heart Association.





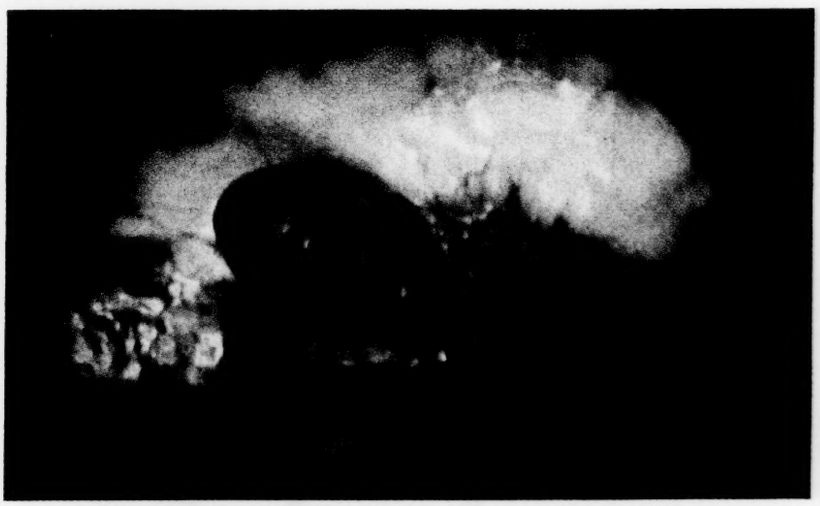


FIG. 1. Photographs taken from cinematographs of the right auricle during auricular flutter. The ectopic focus is near the center of the right auricle. Complete diastole is shown in top illustration. The center and lower figures show the contraction wave passing in all directions from the ectopic focus. If a circus movement were present, the wave would travel in a unidirectional manner.

Auricular Flutter. By Dr. Myron Prinzmetal et al.

The American Journal of Medicine, October, 1951.

minates the arrhythmia. Scherf also found that interruption of the hypothetical circus pathway failed to alter the electrocardiogram of aconitine-produced flutter. Although these observations appeared incompatible with the existence of a circus movement, the methods utilized by all workers during the past thirty years have been too indirect conclusively to prove or disprove Lewis' theory or to establish an alternative hypothesis. Until the course of the flutter wave could be traced throughout all portions of both auricles by direct methods of observation, the mechanism of auricular flutter remained in doubt.

The technics and equipment employed in our laboratory are more direct and more sensitive than those used in previous investigations of auricular flutter. In the experimental animal all portions of both auricles were exposed and the auricular activity recorded on high-speed color cinematographs and direct lead electrocardiograms. In man the electrical activity of the fluttering auricles was studied in esophageal and precordial lead electrocardiograms. By these methods the invalidity of the circus movement theory was established and the entire course of the flutter wave was demonstrated.

EXPERIMENTAL METHODS

The operative technic by which the entire surface of the left and right auricular bodies and appendices were exposed is described in detail elsewhere.[21] After completion of the exposure auricular flutter was produced by (1) electrical stimulation or (2) aconitine application.

1. Flutter was readily obtained during stimulation of various sites on the auricles with single induction shocks from a pulse generator delivered at rates of approximately 400 per minute. Occasionally the arrhythmia persisted a significant length of time after electrical stimulation was discontinued; in such instances cinematographs were recorded during the post-stimulatory as well as the stimulatory phase. All cinematographs of flutter were essentially identical whether recorded during or after the period of stimulation.

2. Local application of aconitine proved to be the most effective method of producing auricular flutter for cinematographic and electrocardiographic study. Initially, 0.05 cc. of an 0.05 per cent solution of the agent was injected into the auricular wall as recommended by Scherf. Subsequently we found that topical application of a 0.05 to 2.0 per cent solution was superior for our purpose. A small cotton swab saturated with the solution was applied to a dried area 1 to 2 mm. in diameter on the surface of the auricle. This modification of Scherf's technic obviates the possibility of accidental injection of the drug into the blood stream and thus reduces the danger of ventricular fibrillation. Flutter was produced most readily by aconitine application to a site on the line of the caudal attachment of the interauricular septum. In every instance in which cinematographs of aconitine-produced flutter were obtained the presence of the arrhythmia was confirmed by simultaneously recorded electrocardiograms. Cinematographs of auricular flutter were identical whether the arrhythmia was produced by electrical stimulation or aconitine application.

Experimentally produced auricular flutter in forty dogs was directly visualized in high-speed cinematographs of the exposed auricles recorded at camera speeds up to 2,000 frames per second. These cinematographs clearly demonstrated for the first time the course of the flutter contraction wave in the auricular appendices and right auricular body. Since the body of the left auricle is immobile, no visual observations could be made in that region. In order to determine the course of the flutter wave in the hitherto unexplored left auricular body and to corroborate the cinematographic observations made in other auricular areas, single and simultaneous direct lead electrocardiograms of experimentally produced flutter were recorded from numerous sites throughout the auricles. Both the cinematographic and electrocardiographic studies establish beyond question that no circus movement is present in auricular flutter.

COURSE OF THE CONTRACTION WAVE OF EXPERIMENTALLY PRODUCED AURICULAR FLUTTER

Regardless of the site of production of the arrhythmia, the contraction wave of auricular flutter arises at the ectopic focus and spreads outward through the contractile auricular musculature in all available directions simultaneously until it reaches the extremities of the auricles. As described elsewhere,[21] identical cinematographic observations on the course of the contraction waves were made in studies of experimental and clinical auricular premature systole and auricular paroxysmal tachycardia. In normal sinus rhythm, also, the contraction

wave spreads outward from its site of origin—in this instance the sino-auricular node—in all available directions simultaneously. Thus the course of the contraction wave is identical in normal sinus rhythm, auricular premature systole, auricular paroxysmal tachycardia and auricular flutter. In none of these rhythms is there evidence of a circus movement, either microscopic or macroscopic. On the contrary, several cinematographically recorded experiments designed to test the validity of Lewis' hypothesis are incompatible with the existence of circus phenomena.

Experiment 1. Auricular flutter was produced by electrical stimulation or aconitine application at the caudal end of the right auricle near the inferior vena cava, the site generally employed by Lewis, and the auricle was photographed at 2,000 frames per second. The contraction wave in the cinematographs was seen to arise at the focus from whence it spread outward through the auricular body and appendix as a broad band involving the entire width of the auricle. Each contraction wave was followed by a relaxation wave arising at the same site and pursuing an identical course. This experiment demonstrates that the contraction wave of auricular flutter does not pursue a narrow pathway, does not turn above the superior vena cava to enter the left auricle and does not circle beneath the inferior vena cava to re-enter its original pathway. Since the contraction wave involves the entire width of the auricle, no daughter waves are present.

Experiment 2. Among the most convincing evidence advanced by Lewis in support of the circus movement theory is a determination of the times of onset of the intrinsic deflections in simultaneously recorded direct leads from the auricles of a dog with experimentally produced flutter. In Lewis' experiment the course of the cardiac impulse was charted by determining its time of arrival at successive points on the auricle between the caudal end and the appendix; occasional observations were made on the left auricular appendix and none on the left auricular body. The time consumed while the impulse traversed the right auricle from the inferior vena cava to the superior vena cava was found to correspond almost exactly with the interval between the arrival of the impulse at the superior cava and its reappearance at the inferior cava. From these calculations Lewis concluded that the impulse traveled at a constant rate of speed up one auricle and down the other to re-enter the original pathway.

In order to check Lewis' computations flutter was produced at a site on the right auricle near the inferior vena cava and the auricle was photographed at 2,000 frames per second. The time in which the contraction wave in the cinematographs traversed the right auricle from the inferior to the superior cava was accurately measured with a stopwatch. Since the length of the return journey down the left auricle along the hypothetical circus pathway is approximately the same as the length of the upward journey through the right auricle, the interval between the onset of succeeding contraction waves should have been about twice the measured interval. In only one of five experiments did the second wave appear at the focus at the anticipated time. In the remaining four instances the interval between the onset of succeeding contraction waves was as much as eight times the measured interval. These results indicate that (1) the onset of the second wave did not represent re-entry of the preceding wave into a circus pathway or (2) the contraction wave traveled through the left auricle as much as four times more slowly than through the right.

Experiment 3. In each of four dogs auricular flutter was produced by stimulation of a site on the caudal attachment of the interauricular septum and both auricular appendices were photographed simultaneously. The cinematographs show that the two appendices usually contracted simultaneously; occasionally, either appendix contracted a fraction of a second before its fellow. According to Lewis' calculations of the course and speed of the flutter impulse the contraction wave should reach the right appendix 0.057 to 0.067 second before its arrival in the left appendix. Such a difference in the actual times of contraction of the two appendices would be magnified to approximately fourteen seconds in the high-speed cinematographs and would be readily apparent to the observer. Since the two appendices in the cinematographs were seen to contract simultaneously or no more than a fraction of a second apart, this experiment proves that the contraction wave of auricular flutter does not pursue a circus pathway up one auricle and down the other.

Experiment 4. In each of three dogs auricular flutter was produced and the hypothetical circus pathway was severed by means of an "inverted

T" burn on the right auricle which was demonstrated cinematographically and electrocardiographically to block effectively the passage of auricular impulses. By repeated stroking with a red-hot glass rod a deep burn was inflicted from the superior to the inferior vena cava, extending well up on the wall of each cava. A second burn was made perpendicular to the first and extending from it across the auricular body to the auriculoventricular groove. Each burn was approximately 1 cm. in width and penetrated the entire thickness of the auricular wall. Cinematographs and simultaneously recorded electrocardiograms revealed that the auricles continued to flutter after the burns were inflicted. If a circus movement as hypothesized by Lewis were present, the inverted T burn would have blocked all possible paths of the impulse as it reached either vena cava and the arrhythmia would have terminated.

Experiment 5. In each of five dogs flutter was produced by application of aconitine to the tip of the right auricular appendix, a site far removed from Lewis' hypothetical circus pathway, and high-speed cinematographs of the right auricle were recorded. The contraction and relaxation waves were seen to initiate at the site of stimulation from whence they traveled across the auricle in a cephalocaudal direction over the appendix and body, terminating near the inferior vena cava. Cooling or amputation of the aconitine focus immediately restored normal sinus rhythm. These observations are incompatible with the circus movement theory for two reasons: (1) Since experimental alteration of a site on the tip of the auricular appendix is capable of initiating and terminating bouts of flutter, the arrhythmia cannot be attributed to the movement of an impulse along a pathway which lies far from the tip of the appendix. (2) If the contraction wave initiating from the ectopic focus on the tip of the appendix pursued a circus pathway, it would travel in a caudocephalic direction to the apex of the appendix on its return journey to the focus. No such returning contraction wave was visible in the cinematographs.

Experiment 6. Auricular flutter was produced from a focus in the center of the right auricle and cinematographs were recorded at 2,000 frames per second. This experiment permitted clear visualization of the flutter contraction wave throughout its entire course. Whether the arrhythmia was produced by aconitine application or as an after-effect of electrical stimulation, the contraction wave arose from the ectopic focus and spread outward in all directions simultaneously, much like ripples on the surface of a pond when a stone is dropped into it. (Fig. 1.) The flutter wave did not follow a unidirectional course as hypothesized in the circus movement theory.

COURSE OF THE EXCITATION WAVE OF EXPERIMENTALLY PRODUCED AURICULAR FLUTTER

In the present study Lewis' electrocardiographic determinations of the course of the flutter wave in the right auricle and left auricular appendix were duplicated and extended. In addition, the flutter wave was traced through the hitherto unexplored left auricular body and the interauricular septum. Simultaneous direct lead tracings from pairs of strategic sites throughout the exposed auricles were recorded by a direct-writing multiple-channel electrocardiograph and interpreted on the basis of two fundamental principles of electrocardiography: (1) The intrinsic deflection in a direct or semi-direct lead is inscribed as the excitation wave passes beneath the recording electrode. Hence the course and speed of the flutter wave could be determined by comparing the times of onset of the intrinsic deflections in the simultaneously recorded tracings. (2) The configuration of the auricular deflections in direct or semi-direct leads varies with the distance between the recording electrode and the site of origin of the excitation wave. The deflections in tracings from electrodes at or near the focus are primarily negative. As the distance between the electrode and the focus increases the positive component of the deflection grows progressively greater in amplitude and duration while the negative component shrinks and eventually disappears. Thus the course of the flutter wave in relation to various sites on the auricles was revealed by differences in configuration of the auricular deflections in the simultaneously recorded tracings. Regardless of the method of electrocardiographic interpretation used, the course of the excitation wave of auricular flutter was found to be identical with the course of the excitation waves of normal sinus rhythm, auricular premature systole and auricular tachycardia, and with the course of the contraction waves of auricular flutter and the slower rate rhythms visualized in the cinematographs. Each wave arises at a single

focus on the auricles and spreads outward through the auricular musculature in all available directions simultaneously. The contraction and excitation waves differ only in that the latter invade all portions of the auricles including the non-contractile body of the left

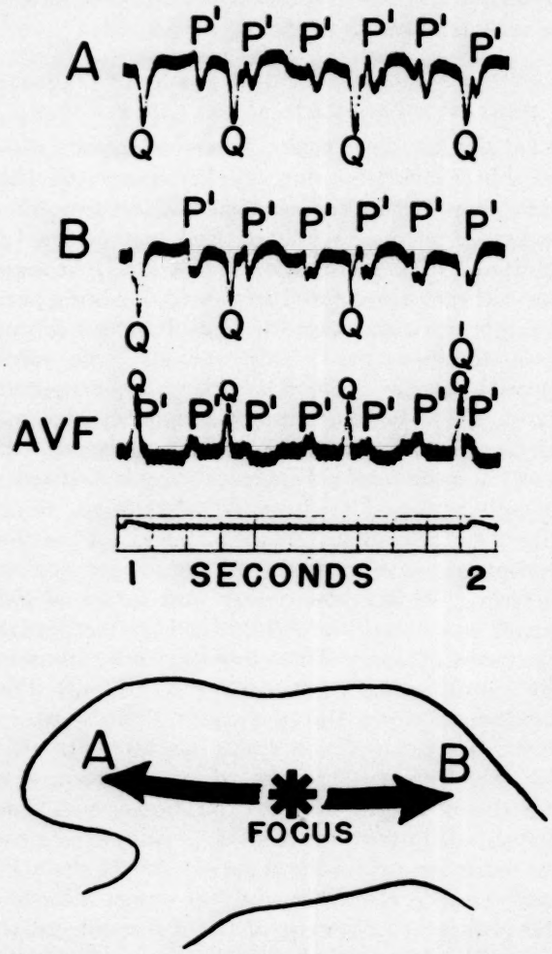


FIG. 2. Direct leads and diagram showing course of excitation wave from an aconitine focus in center of right auricle. Points A and B are equidistant from the focus. The intrinsic deflection from electrodes at points A and B are inscribed simultaneously indicating that the excitation wave spreads from the center of the auricle to both extremities simultaneously.

auricle. As in the cinematographic study, several experiments were performed during the electrocardiographic investigation of experimental auricular flutter which definitely rule out the existence of a circus movement.

Experiment 7. This experiment is the electrocardiographic counterpart of cinematographic experiment 6. In each of nine dogs auricular flutter was produced by application of aconitine to the center of the right auricle while simultaneous electrocardiograms were recorded from two electrodes equidistant from and on opposite sides of the focus. The corresponding intrinsic deflections in the simultaneous tracings were inscribed at virtually the same instant, demonstrating that the excitation wave traveled toward both electrodes simultaneously. (Fig. 2.) Identical results were obtained when flutter was produced by electrical stimulation and electrocardiograms recorded during the post-stimulatory phase of the arrhythmia. If a unidirectional circus movement were present, the excitation wave would have reached one electrode before the other and the intrinsic deflections in the two tracings would not have been recorded simultaneously.

Experiment 8. As noted previously, Lewis produced flutter at a site near the inferior vena cava and timed the onset of the intrinsic deflections in simultaneously recorded leads from the right auricle; from his results he concluded that the flutter wave travels through the right auricle in a caudocephalic direction. This experiment has been duplicated and Lewis' findings in the right auricle confirmed in our laboratory. In order to determine if the excitation wave turns above the superior vena cava to descend through the left auricle in a cephalocaudal direction, as hypothesized by Lewis, a similar experiment was performed while the course of the flutter wave for the first time was traced through both auricles.

I. In each of fifteen dogs flutter was produced from an aconitine focus near the site employed in Lewis' experiment and paired electrodes were placed equidistant from the focus, one on the right auricular body and the other on the left auricular body. The intrinsic deflections in tracings from the paired electrodes were inscribed simultaneously, indicating that the excitation wave reached corresponding sites on the two auricles at the same instant. If the circus movement theory were valid, the wave would not have entered one auricle until it had completed its journey through the other auricle.

II. The aforementioned experiment was repeated while electrocardiograms were recorded from two pairs of electrodes placed at corresponding sites on the bodies of the left and right auricles. Simultaneous tracings from the paired electrodes in each auricle showed that the intrinsic deflections from the electrodes nearest the focus were inscribed before the intrinsic

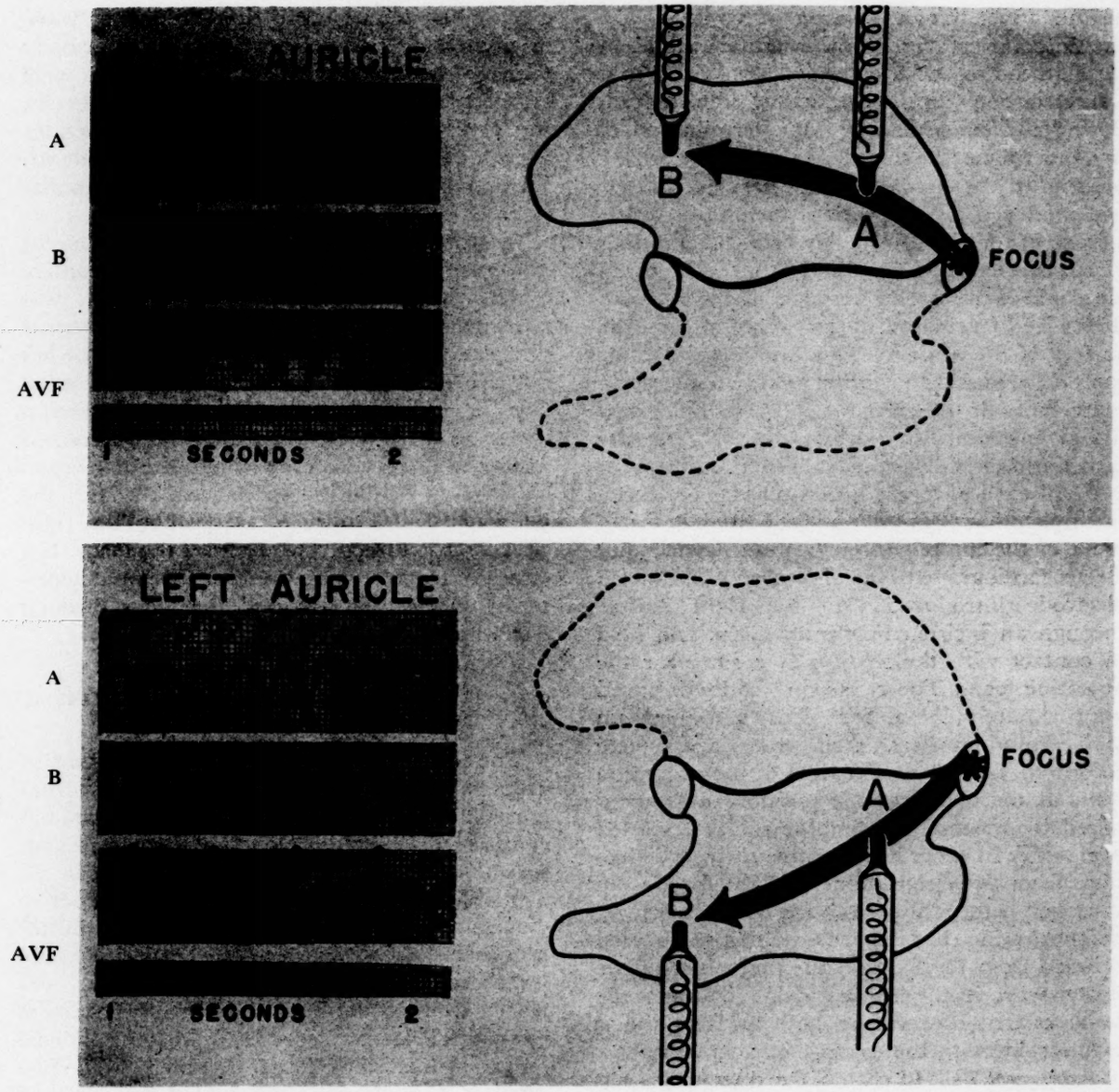


FIG. 3. Direct leads and diagram showing course of excitation wave from an aconitine focus at causal attachment of interauricular septum. In both auricles the intrinsic deflection from electrode B is inscribed after the intrinsic deflection from electrode A. Hence the impulse travels in both auricles in a caudocephalic direction.

deflections from the distal electrodes. (Fig. 3.) It was thus demonstrated that the excitation wave of flutter produced in the caudal region travels through both auricles in a caudocephalic direction. Lewis, in the absence of direct determinations, presumed that the wave traveled through the left auricle in a cephalocaudal direction.

Experiment 9. According to the circus movement theory the flutter wave crosses the interauricular septum twice during each journey around the auricles. Such a movement could not exist unless the septum were incapable of conducting electrical impulses. Otherwise, as the flutter wave passed over the septum it would be transmitted through the septal tissue to adjacent parts of the auricle, the circuit would be interrupted, and the arrhythmia would terminate. The following experiment demonstrates that the role of the interauricular septum in flutter is incompatible with the circus movement theory.

1. In each of four dogs an electrode was introduced into the right auricle through an incision

in the appendix and placed in contact with the interauricular septum. Manual palpation through the right auricular wall confirmed that the electrode tip was touching the septum during the experiment; at the termination of the experiment the auricle was opened and the position of the electrode again verified. Flutter was produced by application of aconitine to the surface of the right auricular body. Continuous electrocardiograms recorded from the septal electrode were in all respects identical with direct lead tracings of flutter recorded from the surface of the auricles. Thus the interauricular septum conducts the flutter wave in the same manner as do other parts of the auricles and would necessarily short-circuit an impulse pursuing Lewis' hypothetical circus pathway.

II. In each of four dogs flutter was produced by application of aconitine at the caudal attachment of the interauricular septum. Continuous electrocardiograms were recorded from an electrode introduced into the right auricle through an incision in the appendix and held in contact with the septum at a site near the aconitine focus. The electrode was then moved approximately 3 cm. cephalad on the septum and continuous electrocardiograms again were recorded. The times of arrival of the excitation wave at the two electrode positions were compared by means of a simultaneously recorded lead AVF. In every instance the intrinsic deflections from the electrode nearest the focus were inscribed before those from the distal electrode, demonstrating that the excitation wave traveled upward from the focus. Thus the course of the flutter wave in the interauricular septum is the same as that observed in both auricles and is inconsistent with the circus movement theory.

Experiment 10. In each of five dogs flutter was produced by application of aconitine to a site between the left and right auricles at the junction of the inferior vena cava and the pulmonary vein. Simultaneous electrocardiograms were recorded from two electrodes on the right auricle, one adjacent to the focus on the auricular body, the other distant from the focus on the appendix. The auricular deflections recorded from the electrode near the focus were primarily negative while the deflections from the electrode on the appendix exhibited a large positive component. Since the positivity of the auricular deflection varies directly with the distance between the recording electrode and the site of origin of the excitation wave, this experiment demonstrates that the flutter wave originated at a point closer to the auricular body than to the appendix. If the flutter wave were self-perpetuating, as hypothesized by Lewis, the configuration of the auricular deflections from all sites on its pathway would be identical since the wave would be without beginning or end.

In order to rule out the possibility of a circus movement which arises anew with each cardiac cycle, the aforementioned experiment was repeated with paired electrodes placed unequal distances from the focus on the body of the left auricle. Again, the auricular deflections recorded from the electrode nearest the focus exhibited a larger negative component than those inscribed from the distal electrode. Thus the excitation wave of auricular flutter travels through the body of the left auricle, as through the right auricle, in a direction away from the focus. If a circus movement were present, the wave necessarily would travel toward the focus through one auricle on its return journey.

COURSE OF THE EXCITATION WAVE OF CLINICAL AURICULAR FLUTTER

The cinematographic and electrocardiographic studies described before establish that auricular flutter in the experimental animal consists of rapidly recurring waves which spread outward from the ectopic focus in all available directions simultaneously. In order to determine if the clinical form of auricular flutter is electrically similar to its experimentally produced counterpart, the flutter wave was traced through the human auricle by means of esophageal leads recorded from ten patients and by precordial leads obtained from two patients. In every instance studied the course of the flutter wave in man was identical with that observed in the experimental animal.

Esophageal Lead Electrocardiogram of Clinical Auricular Flutter

The value of the esophageal lead electrocardiogram derives from the anatomic relationship of the esophagus to the two auricles and the interauricular septum.[22] At levels approximately 20 to 40 cm. from the lips the esophagus lies directly posterior to the auricular structures in sufficient proximity to receive electrical impulses from both the right and the left auricles. Electrodes in the esophagus approximately 35 to

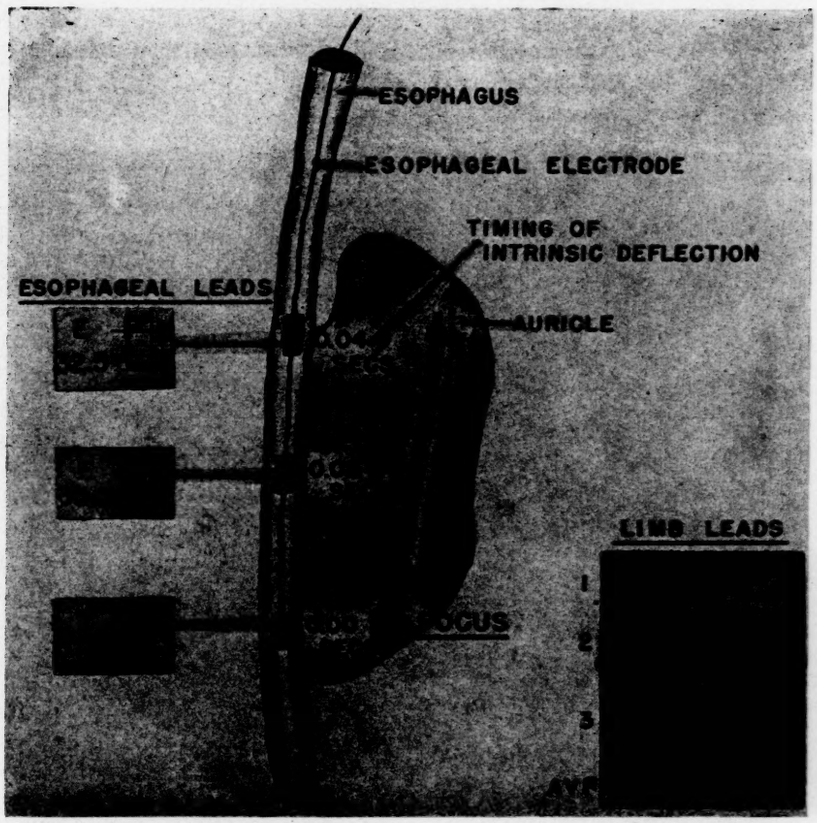


FIG. 4. Esophageal leads and diagram showing course of excitation wave in common type of flutter. The configurations of the deflection from low, mid and high auricular levels indicate that the excitation wave arises in the caudal end of the auricle and travels toward the cephalic end.

40 cm. from the lips record electrical events as they occur in the caudal end of the auricles; electrodes approximately 32.5 to 37.5 cm. from the lips register events occurring in the mid-portion of the auricles; electrodes approximately 30 to 35 cm. from the lips record events in the cephalic end of the auricles. Esophageal lead electrocardiograms are interpreted with reference to the same principles of electrocardiography which provide the basis for the preceding analysis of direct lead tracings from animals. According to these principles a negative deflection is inscribed if the excitation wave is traveling away from the recording electrode, while a positive deflection is inscribed if the wave is approaching the electrode. The intrinsicoid deflection is defined as the sharp negative deflection inscribed when the excitation wave passes directly beneath the recording electrode.

Of the ten instances of clinical auricular flutter studied by means of esophageal leads seven were of the common clinical type exhibiting inverted P′ waves* in limb leads 2, 3 and AVF. Two of the patients manifested the uncommon type of clinical flutter in which the P′ wave is upright in limb leads 1 and 2. The tenth instance represented the rare phenomenon of both the common and uncommon types of flutter occurring at different times in the same patient. As in the experimental animal, the course of the excitation wave of either the common or the uncommon type of auricular flutter in man is incompatible with the existence of a circus movement.

Clinical Observation 1. In each of the seven patients exhibiting the common type of auricular flutter a pure negative auricular deflection consistently was recorded from electrodes in the esophagus directly posterior to or below the

* As will be demonstrated later, a specific portion of the flutter complex in limb leads represents the period of auricular depolarization and is appropriately termed the P′ wave of flutter.

caudal end of the auricles. (Fig. 4.) Esophageal lead tracings from levels directly posterior to or above the cephalic region of the auricles always displayed a pure positive auricular deflection. The auricular deflection in esophageal leads from levels between the caudal and cephalic

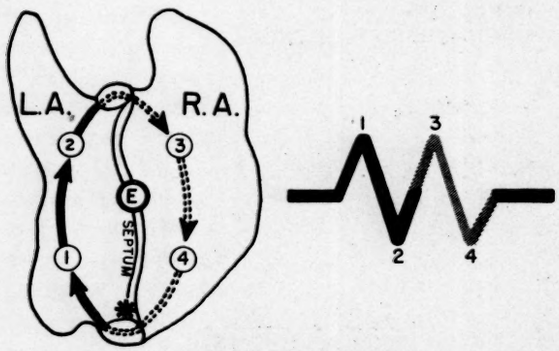


FIG. 5. Hypothetical drawing of M-shaped deflection which would be recorded by esophageal leads from mid-auricular level in common type of auricular flutter, if a circus movement were present. Actually, the shaded portion of the deflection is not recorded; hence no circus movement occurs.

ends of the auricles consisted of a positive wave followed by a negative wave; the amplitude of the positive component varied directly with the distance of the recording electrode from the cephalic extremity. Since a pure negative auricular deflection in semi-direct leads indicates that the excitation wave at all times is traveling away from the recording electrode, the wave in the seven patients with the common type of flutter must have arisen at or near the caudal end of the auricles and traveled toward the cephalic extremity. Conversely, since a pure positive deflection indicates that the wave at all times is approaching the recording electrode, the positive deflection inscribed from the cephalic level of the auricles establishes that the flutter wave traveled toward and terminated at the cephalic extremity. The biphasic deflection recorded from esophageal levels between the caudal and cephalic ends of the auricles provides further evidence that the wave of the common type of flutter travels in a caudocephalic direction; the positive component is inscribed as the wave approaches the mid-auricular region, the intrinsic deflection occurs as the wave passes over the electrode, the negative component is recorded as the wave continues upward to the cephalic extremity. From the previous observations the conclusion may be drawn that the excitation wave of the common type of clinical auricular flutter originates at an ectopic focus in or near the caudal end of the auricles, travels in a caudocephalic direction and terminates at the cephalic end of the auricles. This conclusion correlates with two experimental observations noted previously: (1) Flutter in the dog is most easily produced from sites in the caudal region of the auricles and (2) the excitation wave of auricular flutter produced in the caudal region travels through both auricles in a caudocephalic direction. Thus the region most prone to give rise to auricular flutter, as well as the course of the flutter excitation wave originating in this region, are the same in man as in the experimental animal.

As pointed out earlier, the fundamental principles of electrocardiography imply that the auricular deflections in direct leads recorded from all sites on an unbroken circuit would be identical in configuration. Since these principles are applicable to esophageal as well as direct lead electrocardiograms, it follows that esophageal leads from all sites posterior to a circus pathway would exhibit identical auricular deflections if the excitation wave traversing the pathway were of the self-perpetuating type hypothesized by Lewis. Thus the observed variation in configuration of the flutter deflections recorded from different levels of the esophagus in man is incompatible with the existence of a self-perpetuating circus movement. The tracings from the seven patients described before also rule out the possibility of a non-self-perpetuating circus movement, as shown in the following hypothetical description of the esophageal lead electrocardiogram which would be recorded if the excitation wave of the common type of clinical flutter pursued a circus pathway.

Assuming a circus movement is present in the common type of flutter, an electrode in the esophagus at the level of the focus in the caudal end of the auricles would record a negative deflection as the excitation wave traveled through one auricle in a caudocephalic direction until it reached the cephalic extremity. Since electrical events in both auricles are recorded from the esophagus, this negative deflection would be followed by a positive deflection representing the return journey of the wave during which it is presumed to travel down the other auricle until it arrives at the focus. In neither the present nor previous studies has such a positive component been recorded in

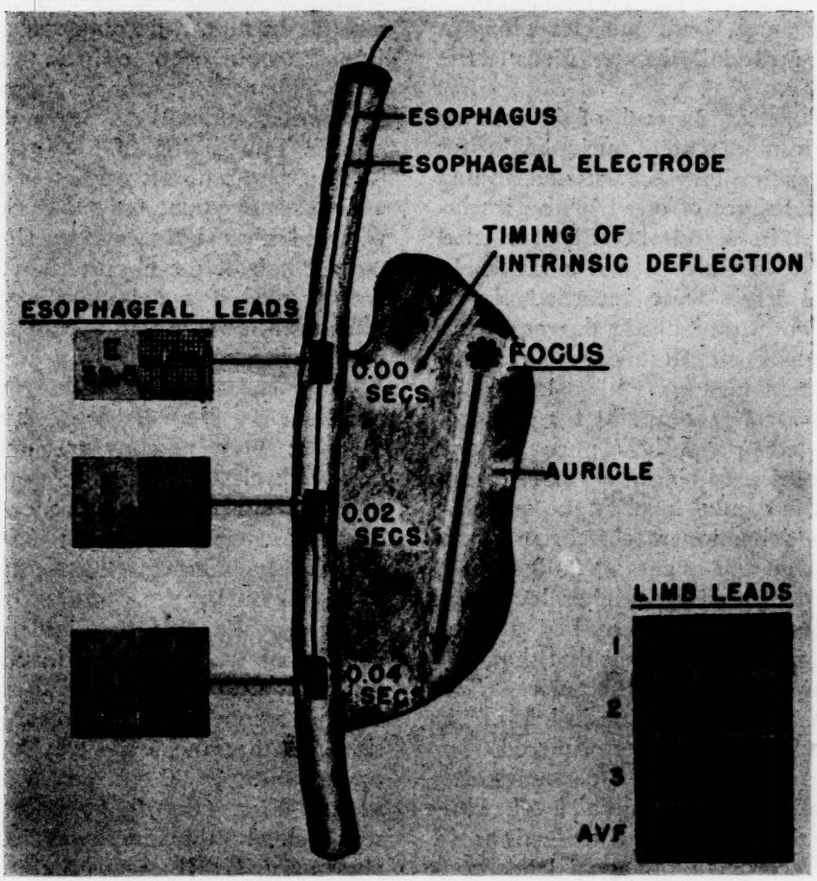


FIG. 6. Course of the excitation wave in the uncommon type of clinical auricular flutter. The configuration of the deflections from various levels of the auricles indicates that the wave arises in the cephalic region and travels in a cephalo-caudal direction.

esophageal leads from the caudal level of the auricles, demonstrating that the flutter wave does not return to the focus via a circus pathway. Again assuming a circus movement is present in the common type of flutter, an electrode in the esophagus at the mid-auricular level would register a positive deflection as the excitation wave traveled from the caudal focus toward the mid-portion of one auricle, then a negative deflection as the wave continued through the same auricle in a direction away from the electrode until it reached the cephalic end. This biphasic deflection would be followed by a second biphasic deflection, representing the journey of the wave through the other auricle as it traversed the third and fourth quadrants of the circuit on its return to the focus. (Fig. 5.) Actually, the second of this hypothetical series of two positive-negative deflections is not inscribed; hence the second half of the circuit is non-existent. Once again assuming a circus movement is present, an esophageal lead tracing of the common type of flutter recorded from the cephalic level of the auricles would exhibit a positive deflection recorded during the outgoing journey of the excitation wave up one auricle from the focus to the cephalic end, followed by a negative component representing the course of the wave through the other auricle as it recedes from the electrode on its return journey to the focus. In fact, no negative component is visible in esophogeal leads from the caudal level of the auricles, indicating that the excitation wave terminates at the cephalic extremity. It is thus apparent that esophageal lead electrocardiograms of the common type of clinical flutter, whether recorded from caudal, cephalic or middle auricular levels, exhibit none of the complex deflections which necessarily would be present if a circus movement occurred. Rather, the configuration of the deflections establishes that the excitation wave of the common type of

flutter travels through both auricles along a linear path from a caudally located focus to the cephalic extremity.

Clinical Observation 2. In each of six patients exhibiting the common type of auricular flutter the course of the excitation wave was determined by comparing the times of onset of the intrinsicoid deflections in simultaneously recorded esophageal leads from different auricular levels. Four esophageal leads were recorded simultaneously in two of the patients. In the remaining four instances three simultaneous esophageal leads were obtained together with a lead AVF which served as a reference point for purposes of timing. As noted previously, a pure negative deflection was consistently inscribed in esophageal leads from the caudal level of the auricles; the onset of this deflection marks the origin of the excitation wave and occurs at 0.00 second. In tracings from successively higher levels the intrinsicoid deflections were inscribed at progressively later periods, indicating that the wave was traveling upward through the auricles. The onset of the intrinsicoid deflection in leads from the cephalic level of the auricles occurred from 0.04 to 0.06 second later than the corresponding deflection in leads from the caudal level. Thus the timing of the intrinsicoid deflections in simultaneously recorded esophageal leads confirms the conclusion derived from an analysis of the configuration of the auricular deflections in esophageal leads: Both methods demonstrate that the excitation wave of the common type of clinical flutter travels from a focus at the caudal end of the auricles in a caudocephalic direction until it terminates at the cephalic extremity.

Clinical Observation 3. Lewis' hypothetical circus pathway covers the caudocephalic length of one auricle plus the cephalocaudal length of the other auricle. Hence the total distance traveled by an excitation wave pursuing the pathway would equal twice the caudocephalic length of the auricles. As noted before, in seven instances of the common type of flutter the excitation wave was found to arrive at the cephalic end of the auricles 0.04 to 0.06 second after its departure from the focus in the caudal extremity. If the wave were engaged in a self-perpetuating circus movement at a constant rate of speed, presumably it would complete the entire circuit in 0.08 to 0.12 second. In all seven patients, however, the interval between the onset of succeeding cardiac cycles (the P′-P′ interval) was appreciably longer than 0.12 second. In fact, a cardiac cycle of 0.08 to 0.12 second would occur only if the auricular rate were 500 to 750 beats per minute, yet clinical fibrillation generally supervenes at far slower rates.[21] The timing of the intrinsic deflections in the seven patients studied is compatible with a circus movement only if the excitation wave traveled several times more slowly during the second half of the circuit than during the first half or/and if a delay occurred between the return of the wave to the focus and its departure on the succeeding journey around the pathway.

The speed of an excitation wave in the human auricle may be calculated by the following method: Simultaneous tracings are recorded from two electrodes in the esophagus a known distance apart. The difference in the time of onset of corresponding intrinsic deflections in the simultaneous tracings is then determined and divided by the distance between the two electrodes. By this method the speed of the excitation wave in an instance of clinical auricular flutter was found to approximate 125 cm. per second; in a human subject with normal sinus rhythm the wave traveled at a speed of approximately 500 cm. per second. The experimental observation that the flutter wave travels more slowly than the wave of normal sinus rhythm originally was made by Lewis in a direct lead study of dogs and has been confirmed cinematographically.

Clinical Observation 4. Single and simultaneous esophageal leads from various auricular levels were recorded from each of two patients exhibiting the uncommon type of auricular flutter in which the P′ waves in limb leads 1 and 2 are directed upward. In both instances the direction of the deflections in esophageal leads from each auricular level as well as the comparative times of onset of the intrinsicoid deflections in simultaneous tracings from different levels were exactly opposite those observed in the common type of flutter. The auricular deflections in esophageal leads from the cephalic level were completely negative while those from the caudal level were entirely positive (Figure 5), indicating that the excitation wave arose in the cephalic end of the auricles and terminated in the caudal region. The intrinsicoid deflections in simultaneous leads from progressively lower levels were recorded at successively later periods, again indicating that the excitation wave traveled in a cephalocaudal direction. From both the configuration and the timing of the

auricular deflections in esophageal leads, therefore, the conclusion may be drawn that the essential difference between the common and uncommon types of auricular flutter lies in the location of the ectopic focus. The common type of flutter arises from a focus in the caudal region; the uncommon type originates in the cephalic region. In both types the excitation wave pursues a linear course from the focus to the opposite extremity of the auricles where it terminates. In neither type is there a circus movement.

Clinical Observation 5. One patient observed during the study manifested both the common and uncommon types of flutter described before. On some occasions the limb lead electrocardiogram of this patient exhibited the common type of flutter with inverted P′ waves in leads 2, 3 and AVF; esophageal leads recorded on such occasions showed pure negative deflections from the caudal level of the auricles and pure positive deflections from the cephalic level, indicating that the excitation wave arose at the caudal extremity and pursued a caudocephalic course. On another occasion limb lead tracings from the same patient were characteristic of the uncommon type of flutter with upright P′ waves in leads 1 and 2; the deflections in esophageal leads from the cephalic level were completely negative while those from the caudal level were completely positive, demonstrating that the ectopic focus was then at the cephalic end of the auricles and the excitation wave traveled in a cephalocaudal direction. The auricular rate was identical whether the common or the uncommon type of flutter prevailed.

Precordial Lead Electrocardiogram of Clinical Auricular Flutter

In two patients exhibiting the common type of auricular flutter the intrinsicoid deflections in precordial leads were sufficiently distinct to permit definite identification. The course of the flutter wave was charted on the chests of these patients by comparing the times of onset of the intrinsicoid deflections in multiple precordial leads with reference to a simultaneously recorded lead 3 or AVF. In each instance a pure negative deflection was recorded in precordial leads from the lower end of the sternum indicating that the ectopic focus (time point 0.00) was at the caudal end of the auricles. The time values of the intrinsicoid deflections in leads from all sites over both auricles varied directly with the distance between the recording electrode and the focus; that is, corresponding deflections in leads from sites equidistant from the focus were inscribed at the same time point. These observations establish that the excitation wave traveled outward and upward from the focus in all available directions, reaching sites on both auricles equidistant from the focus at approximately the same time. If a circus movement were present, the excitation wave would complete its course through one auricle before entering the other and would travel toward the focus throughout the last half of the circuit; consequently, the time values of the intrinsicoid deflections in precordial leads from all sites over one auricle would be greater than those from all sites over the other auricle, and the values of the deflections recorded from either the left or the right auricle would vary inversely with the distance of the electrode from the focus. Thus the course of the excitation wave of auricular flutter as traced by precordial leads in man confirms the observations made by means of esophageal leads in man and by direct leads and cinematographic visualization in the experimental animal: The flutter wave spreads outward from the ectopic focus through both auricles simultaneously. It does not pursue a circus pathway.

LIMB LEAD ELECTROCARDIOGRAM OF AURICULAR FLUTTER

By the methods of direct and semi-direct lead electrocardiography described previously, the fact was established that the course of electrical events in the auricles of man and experimental animal during auricular flutter is essentially similar to that occurring during normal sinus rhythm, auricular premature systole and auricular paroxysmal tachycardia. In each rhythm the excitation waves arise from a single site and spread outward through both auricles in all available directions simultaneously. One might expect, therefore, that the auricular complexes in limb leads, which are graphic representations of electrical events in the auricles, also would be essentially similar in all four rhythms. The resemblance between the P wave of normal sinus rhythm and the P′ waves of auricular premature systole and paroxysmal tachycardia is well known; differences in configuration of these deflections have been demonstrated to result from differences in location of the initiating focus. The undulating "F" wave of flutter, how-

ever, is considered specific for the arrhythmia and is generally believed to represent the movement of the excitation wave along a circular pathway entirely unrelated to the path pursued by the waves of the slower rate rhythms. Indeed, the only evidence advanced to establish the existence of a circus movement in clinical flutter is derived from an examination of the momentary atrial electrical axes of the flutter undulation. From such an analysis Lewis concluded, "This axis revolves during the progress of each auricular cycle through 360 degrees. This revolution gives us, so we believe, incontestable evidence that the movement of the excitation wave throughout the auricle as a whole is controlled by a re-entrant movement around a circle or an ellipse."[23] Dechard, Ruskin and Hermann arrived at similar conclusions after repeating Lewis' experiment with certain modifications.[24]

The normal P wave represents depolarization of the auricular musculature and is recorded during the interval between the onset and termination of the excitation wave. A Ta segment, representing auricular repolarization, sometimes appears following the P wave but generally is of insignificant amplitude or is masked by the ventricular complex. If no ventricular complex interferes, as in the presence of auriculoventricular block, a small Ta wave is inscribed in a direction opposite that of the P wave in about 50 per cent of cases. The following experiments are designed to demonstrate that the auricular complex of flutter, like the ventricular complex, consists of two components; namely, a wave of depolarization followed by a wave of repolarization. The first of these components represents the period of excitation in the fluttering auricle and is an exact counterpart of the P wave of normal sinus rhythm and the P′ waves of auricular premature systole and auricular paroxysmal tachycardia. The second component, the Ta segment, is rendered visible by the rapidity of the auricular rate coupled with the presence of auriculoventricular block. In the absence of the Ta segment the auricular complex of flutter is essentially identical with that of the slower-rate rhythms.

P′ Wave of Auricular Flutter

Experiment 1A. Flutter was produced by application of aconitine to the extreme caudal end of the auricle over the interauricular septum. Two electrodes were placed on the same auricle, one at the caudal extremity close to the focus and the other at the tip of the appendix as far removed as possible from the focus. Electrocardiograms from the two electrodes were recorded simultaneously with lead AVF. As discussed earlier, the intrinsic deflection in the direct lead from the region of the focus is inscribed during or immediately after the onset of the excitation wave; the intrinsic deflection in the lead from the opposite end of the auricle is recorded at or immediately preceding the termination of the wave. Hence the portion of the simultaneous limb lead inscribed during the interval between the onset of the intrinsic deflections in the two direct leads must represent the period of auricular excitation. When the direct leads were projected on lead AVF the intrinsicoid deflections in the tracing from the focal area were found to correspond to the beginning of the descending limb of the trough of the flutter complex in the limb lead. The intrinsic deflections in the tracing from the electrode distal to the focus usually corresponded to the function of the ascending limb of the trough with the upward bowing of the flutter complex in the limb lead. Similar results were obtained when the experiment was repeated with the two electrodes placed directly on the septum at opposite ends of the auricle. It was thus demonstrated in the experimental animal that a specific limited portion of the flutter complex in limb leads represents the entire period of auricular excitation.

Experiment 1B. In each of ten instances of clinical flutter a limb lead was recorded simultaneously with two esophageal or precordial leads from opposite ends of the auricle. When the semi-direct lead tracings were projected on the limb lead the portion of the flutter complex in the limb lead, which corresponded to the period of auricular excitation, was identical with that delineated in experiment 1A.

The preceding experiment establishes that the period of auricular excitation in both clinical and experimentally produced auricular flutter is represented in limb lead electrocardiograms by a specific portion of the auricular complex. Since the term "P wave" or "P′ wave" is used to describe the electrocardiographic manifestations of auricular excitation in normal sinus rhythm, auricular premature systole and auricular paroxysmal tachycardia, consistency would demand that similar terminology be applied to the electrocardiogram of auricular flutter. For this reason it is proposed that the specified por-

tion of the flutter complex in limb leads be referred to as the P′ wave of auricular flutter.

The demonstration that only a limited portion of the flutter complex represents auricular excitation invalidates Lewis' analysis of the momentary atrial electrical axis in the limb lead electrocardiogram of clinical flutter. Lewis' conclusions were drawn from a determination of the electrical axis of the *entire* auricular complex which he assumed to represent the course of the excitation wave. When the momentary atrial electrical axis of only the P′ portion of the auricular complex is calculated, the axis is found to revolve through less than 360 degrees and consequently is incompatible with the existence of a circus movement.

Experiment 2. Auricular tachycardia was produced by application of aconitine to the body of the right auricle near the inferior vena cava and continuous simultaneous limb lead electrocardiograms were recorded. As the rate of discharge from the aconitine focus gradually increased the tracings displayed a complete series of patterns ranging from the sharp deflections characteristic of tachycardia to the typical sawtooth undulations of flutter. At the onset of the tachycardia a sharply inverted P′ wave followed by a distinct isoelectric period was present in lead 3. With the gradual increase in auricular rate the configuration of the P′ wave remained constant but the P′-R interval lengthened, an upright Ta wave appeared and 2:1 auriculoventricular block developed. These changes were sufficient to produce the undulatory appearance of flutter; the P′ portion of the flutter complex was virtually indistinguishable from the P′ wave recorded throughout the episode of tachycardia. When a transition from flutter to tachycardia was produced by cooling the aconitine focus, the reverse of the gradual changes described before was seen to occur. Again the configuration of the P′ wave remained constant while progressive alterations in the remaining portions of the electrocardiogram produced all possible patterns intermediate between the undulations of flutter and the sharp deflections of tachycardia. When both disorders arise from the same ectopic focus, therefore, the P′ wave of tachycardia and the P′ portion of the flutter complex are similar in configuration.

The clinical counterpart of experiment 2 occurs when a single patient exhibits paroxysms of auricular tachycardia and bouts of flutter arising at different times from the same ectopic focus. Such unusual instances have been observed in the present study as well as by other workers.[25–28] As in the experimental animal, the P′ wave inscribed during the paroxysms of clinical tachycardia is similar in contour to the P′ portion of the auricular complex recorded during episodes of flutter from the same focus.

Experiment 3. The transition from tachycardia to flutter was initiated by application of aconitine to the tip of the right auricular appendix while limb leads 1, 2 and 3 were recorded simultaneously. The aconitine focus on the appendix was then frozen with ethyl-chloride spray which abolished its pacemaking capacity for the duration of the experiment; normal sinus rhythm supervened. A new focus was produced by application of aconitine to the caudal end of the right auricle in the same animal; again the transition from tachycardia to flutter was recorded in leads 1, 2 and 3. The electrocardiograms taken while the ectopic focus was located on the appendix exhibited upright P′ waves of similar configuration throughout the transition from tachycardia to flutter. In the tracings recorded while the ectopic focus lay in the caudal extremity of the same auricle, the P′ wave inscribed during both tachycardia and flutter was sharply inverted in leads 2 and 3. Thus the configuration of the P′ waves of tachycardia and flutter varies in similar manner with the location of the ectopic focus.

The preceding three experiments demonstrate that the auricular complex of flutter in man and experimental animal includes a component which represents the period of auricular excitation and may properly be called the P′ wave of flutter; that the P′ component in the flutter complex is similar in configuration to the P′ wave of tachycardia when the two arrhythmias arise from the same ectopic focus; that the P′ waves of both tachycardia and flutter vary in contour with the location of the ectopic focus. These observations may prove of clinical value since the configuration of the P′ wave in any given instance of either tachycardia or flutter reveals the region of the auricle from which the arrhythmia initiates. Similarly, any change in configuration of the P′ wave of either tachycardia or flutter represents a change in location of the ectopic focus.

Ta Wave of Auricular Flutter

As described previously, the electrocardiographic changes observed in limb leads during the transition from auricular tachycardia to

auricular flutter consist of the emergence of a second component of the auricular complex which gradually replaces the isoelectric interval between the P′ wave and the ventricular complex. This second component is inscribed in a direction opposite that of the P′ wave, lending

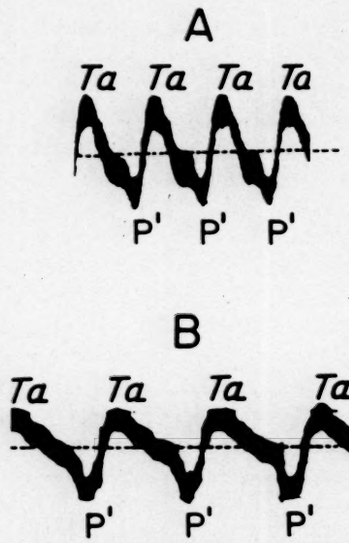


FIG. 7. P′ waves followed with oppositely directed Ta waves. A, auricular rate is 400 per minute followed with very slight isoelectric shelf; B, slower auricular rate, the isoelectric shelf is broader and the Ta wave lower in amplitude.

to the electrocardiogram the familiar undulatory appearance of flutter. (Figs. 7 and 8.)

In both man and experimental animal one of the factors which determines the size of the Ta wave is the auricular rate. During relatively slow tachycardia the P′ wave is followed by an isoelectric period. (Fig. 9.) At more rapid auricular rates the second limb of the P′ wave extends beyond the isoelectric line to form the first limb of the Ta wave; the second limb of the Ta wave descends in a gentle slope back to the isoelectric line. With further increases in auricular rate the Ta wave becomes larger and more bowed until eventually it may usurp the entire space formerly occupied by the isoelectric period; the second limb of the Ta wave then joins the first limb of the P′ wave to form the continuous undulatory pattern of flutter of the pure sine wave type.

When the auricular rate is relatively slow or when the excitation wave arises near the sinus node, limb lead electrocardiograms often are recorded which fulfill all other criteria for a diagnosis of auricular flutter but exhibit little or

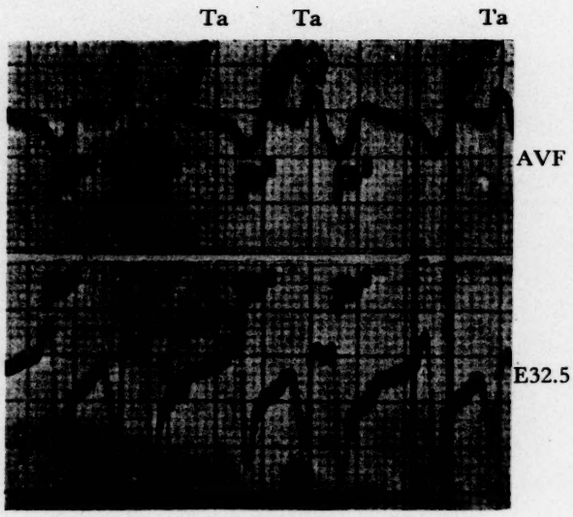


FIG. 8. Simultaneously recorded esophageal and AVF leads. By projecting esophageal lead on AVF, period of depolarization (P′ wave) is delineated. The remaining portion of the limb lead must represent auricular repolarization and is the Ta wave.

no Ta wave. (Fig. 10.) Conversely, in infants and less frequently in adults, tracings of rapid-rate arrhythmias with 1:1 ventricular response may display large Ta waves which combine with the P′ waves to produce an undulatory pattern. A study of tracings from patients with complete auriculoventricular dissociation coupled with either auricular tachycardia or flutter has shown that Ta waves may be present in one limb lead and absent in a simultaneously recorded lead. Whether such instances should be diagnosed as tachycardia or flutter is a subject of controversy.

The foregoing observations indicate that the Ta wave of auricular flutter is induced or rendered visible by the rapidity of the auricular rate. Cinematographic study of the auricles during rapid-rate tachycardia and flutter has shown that the period of diastole in these arrhythmias is extremely brief and the auricular musculature has little time to rest between succeeding contractions; as a result of overwork, the auricles are strained. If biochemical changes are associated with mechanical and electrical events, it seems possible that catabolites would accumulate in the rapidly beating auricles which would affect the process of auricular repolarization. Thus the Ta wave recorded at rapid auricular rates may be related to auricular strain due to overwork. Certainly an analogy to the T wave in the electrocardiographic pattern of ventricular strain is suggested by the fact that both the

T wave and the Ta wave are inscribed in a direction opposite that of the preceding deflection. Final elucidation of the effect of auricular injury and of auricular strain on the formation of Ta waves must await further study.

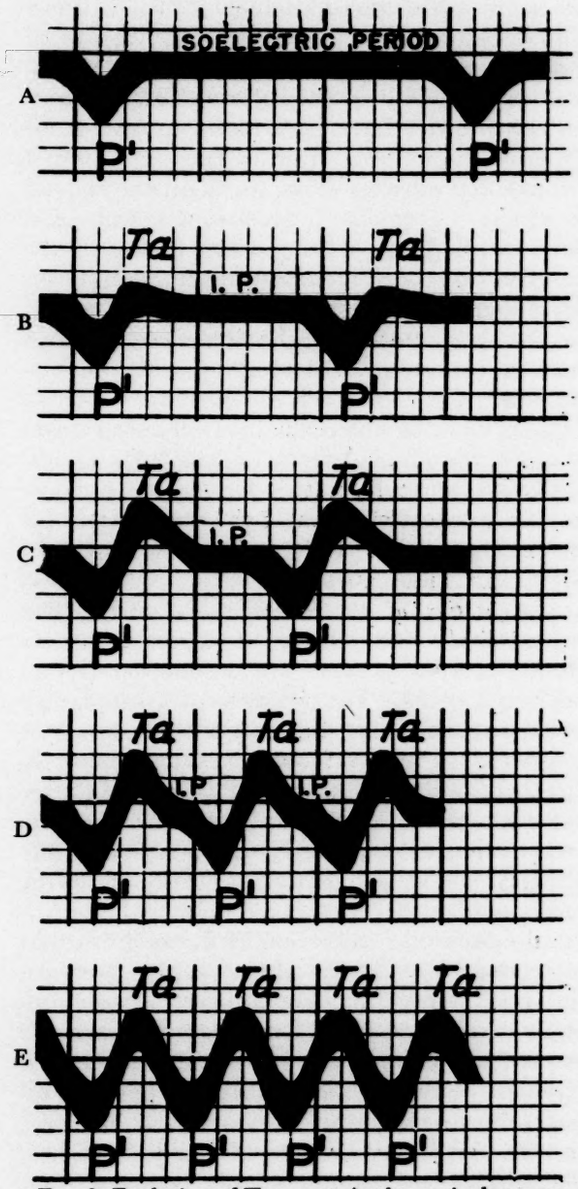


FIG. 9. Evolution of Ta wave. As the auricular rate increases, the isoelectric period is gradually replaced by a Ta wave until typical undulating pattern of flutter is present.

SITE OF THE ECTOPIC FOCUS IN CLINICAL AURICULAR TACHYCARDIA AND AURICULAR FLUTTER

Innumerable experiments in animals as well as esophageal lead studies in man have revealed that an inverted P′ wave is recorded in limb leads 2 and 3 when the excitation wave originates in the caudal region of the auricles, while an upright P′ wave in leads 1 and 2 represents an excitation wave arising in the cephalic region. In

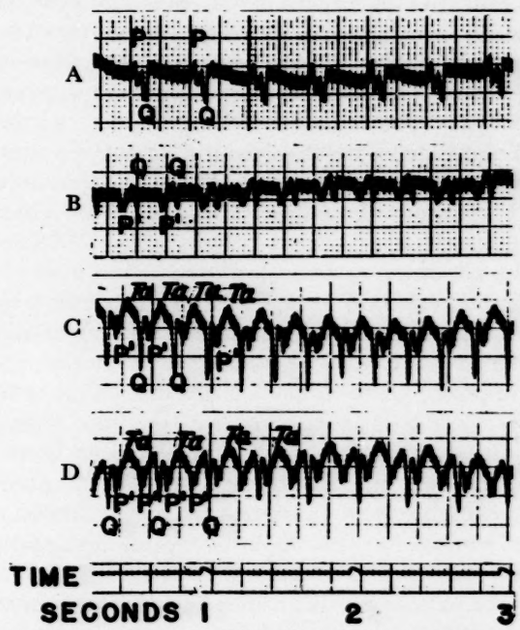


FIG. 10. Electrocardiogram from animal with ectopic focus at caudal end of auricle, illustrating relationship between auricular rate and size of Ta wave. A, sinus tachycardia—auricular rate, 158 per minute; B, paroxysmal auricular tachycardia —auricular rate, 220 per minute; C, paroxysmal auricular tachycardia—auricular rate, 264 per minute; D, auricular flutter—auricular rate, 375 per minute.

order to determine the most frequent sites of origin of the clinical arrhythmias an analysis was made of the configuration of the P′ waves in limb lead electrocardiograms from 139 patients with auricular flutter and 107 patients with auricular tachycardia. Of the 139 examples of flutter almost 70 per cent exhibited deeply inverted P′ waves in leads 2 and 3, indicating that the focus was in the caudal region; such instances represent the common or caudal type of flutter. In 15 per cent of the tracings of flutter the P′ wave was upright in leads 1 and 2, implicating the cephalic region as the site of origin of the disturbance; these instances are of the uncommon, or cephalic, type of flutter. In the remaining 15 per cent of instances of flutter the P′ waves could not be identified in limb leads; under such circumstances the location of the ectopic focus is indeterminable. Of the 107 examples of

clinical tachycardia less than 10 per cent displayed the inverted P′ wave in limb leads indicative of a focus in the caudal region, while 29 per cent presented the upright deflection characteristic of the caudal type of arrhythmia. The remaining 62 per cent of instances of tachycardia were of the indeterminate type in which little or no evidence of auricular activity is visible in limb leads. Probably the majority of tachycardias assigned to the indeterminate group are of the cephalic type, since the upright P′ waves associated with a cephalic site of origin are more readily obscured by the preceding ventricular complex than are the sharply inverted P′ waves of the caudal type of arrhythmia.

The aforementioned statistical analysis suggests that auricular tachycardia and auricular flutter usually, although not always, originate in opposite extremities of the auricles. The majority of instances of flutter arise in the caudal region while only a small minority start in the cephalic region. Insofar as its site of origin is determinable, tachycardia appears to exhibit a predilection for the cephalic region and only rarely arises in the caudal region. If such a fundamental anatomic difference generally exists between tachycardia and flutter, it may account for certain of the clinical dissimilarities of the two arrhythmias.

RELATIONSHIP BETWEEN AURICULAR TACHYCARDIA AND AURICULAR FLUTTER

By means of high-speed cinematography and direct lead electrocardiography the unitary nature of auricular tachycardia and auricular flutter in the experimental animal has been demonstrated. Both arrhythmias may be produced by stimulation of a single site on the auricle; both arrhythmias terminate when the site of stimulation is inactivated. In both arrhythmias the contraction and excitation waves arise from the focus in an identical manner and pursue an identical course through the auricles; in neither arrhythmia is there a circus movement. As the auricular rate is progressively increased the mechanical and electrical characteristics of tachycardia gradually disappear and the characteristics of flutter gradually emerge; as the auricular rate is progressively decreased, the reverse of these changes gradually occurs. From this series of observations the conclusion is drawn that auricular tachycardia and auricular flutter in the experimental animal are manifestations of the same fundamental disturbance, identical in mechanism and differing essentially only in rate.

Because of limitations of technic equally conclusive evidence of the unitary nature of auricular tachycardia and auricular flutter in man cannot be obtained. Nevertheless, by means of semi-direct lead electrocardiography the excitation waves of clinical auricular tachycardia and auricular flutter have been found to arise from a single ectopic focus, from whence they spread through the auricles along an identical course; no circus movement is present. In limb lead electrocardiograms a component of the flutter complex has been demonstrated which is an exact counterpart of the P′ wave of tachycardia; the remaining portion of the flutter complex has been shown to depend largely upon the auricular rate. This series of observations strongly suggests that no difference in mechanism exists between the clinical forms of auricular tachycardia and auricular flutter. Under such circumstances a re-examination of the various criteria currently used to differentiate the two arrhythmias is indicated.

Configuration of the Auricular Deflection. The typical limb lead tracing of auricular tachycardia consists of a P′ wave followed by an isoelectric period. The characteristic undulatory pattern of auricular flutter has been shown to consist of a P′ wave followed by an oppositely directed Ta wave. Since the P′ waves of flutter and tachycardia are similar in contour when the arrhythmias arise from the same region of the auricles, as is frequently the case, only the presence of the Ta wave could serve to differentiate the auricular deflection of flutter from that of tachycardia. During the transition between the two arrhythmias replacement of the isoelectric period by the Ta wave occurs gradually as the auricular rate increases; the precise moment at which the undulatory pattern appears must be arbitrarily defined. Furthermore, the Ta wave may be present in one limb lead and absent in a simultaneously recorded lead; yet such electrocardiograms could not represent the occurrence of two distinct arrhythmias in the same auricle at the same time. It is thus apparent that the configuration of the auricular deflection does not provide a basis for differentiating between auricular tachycardia and auricular flutter in borderline cases nor does it constitute evidence that the mechanism of the two arrhythmias is different.

Sine Wave. A pure sine wave is formed when the Ta wave of flutter is sufficiently broad to encompass the entire interval between the P′ wave and the succeeding auricular complex. The majority of electrocardiograms now diagnosed as flutter exhibit an isoelectric interval or "shelf" following the Ta wave. Since the breadth of the Ta wave varies directly with the auricular rate, a sine wave is recorded only when the flutter rate is extremely rapid or when the configuration of the auricular deflection is influenced by medication. Hence the presence of a sine wave cannot be considered an essential characteristic of auricular flutter.

Auriculoventricular Block. Physiologic auriculoventricular block must be differentiated from block due to pathologic interruption of the auriculoventricular conduction system. Physiologic block occurs when the auricles beat so rapidly that the ventricles are unable to respond to each auricular contraction; hence the presence of this phenomenon depends upon the auricular rate and the efficiency of the auriculoventricular node and conduction system. Since the Ta wave also is precipitated by rapid auricular rates, physiologic auriculoventricular block is present in most instances displaying the characteristic undulatory pattern of flutter. Pathologic auriculoventricular block may occur at any auricular rate and occasionally is seen in normal sinus rhythm as well as in all auricular arrhythmias. Thus the presence of auriculoventricular block does not differentiate flutter from tachycardia and does not represent a difference in mechanism of the two arrhythmias.

Effect of Vagal Stimulation. Vagal stimulation, whether effected by carotid sinus massage or by vagus-stimulating drugs, frequently terminates slow auricular paroxysmal tachycardia but rarely stops rapid tachycardias and tends to convert flutter to fibrillation. Lewis[29] observed and we have confirmed that this difference in response is related to auricular rate; in the experimental animal vagal stimulation causes asystole if applied while the auricles are beating more slowly than a certain critical rate but is ineffective or precipitates fibrillation if applied while the auricles are beating more rapidly than the critical rate. Hence the response of tachycardia and flutter to vagal stimulation varies with the auricular rate rather than with the mechanism of the arrhythmia.

Auricular Rate. The noteworthy feature shared by the various criteria discussed before is that each bears a direct relationship to auricular rate. When the auricular rate is between the rate of discharge from the sinus node and the fibrillation threshold, either tachycardia or flutter prevails. Within this rate range the more rapid the auricular rate the greater is the likelihood that the characteristics of flutter—undulatory pattern, Ta wave, auriculoventricular block, increase in auricular rate during vagal stimulation—will be present. Conversely, the characteristics of tachycardia—P′ wave, isoelectric period, 1:1 ventricular response, termination by vagal stimulation—are found with increasing frequency as the auricular rate declines from the fibrillation threshold. No specific range of auricular rates serves to differentiate flutter from tachycardia; the various characteristics of each arrhythmia develop at different auricular rates in the same subject, and the rate at which a given characteristic appears is variable from subject to subject and probably in the same subject at different times. It is thus apparent that no line of demarcation exists between tachycardia and flutter. Rather, flutter is a form of auricular tachycardia usually originating at the caudal end of the auricle and occurring at a rate sufficiently rapid to precipitate physiologic auriculoventricular block.

The confusion which prevails among electrocardiographers concerning the diagnosis of tachycardia and flutter is understandable in view of the previous observations. Only by invoking arbitrary criteria can the two arrhythmias be differentiated in borderline cases. Every electrocardiographic department has encountered tracings which exhibit some but not all of the characteristics of flutter; the diagnosis in these instances varies with the particular criteria employed. In order to avoid such futile controversy we have found it expedient to apply the term auricular tachycardia to all auricular arrhythmias now classified as flutter or tachycardia, and to supplement this general diagnostic term with a description of the physiologic characteristics of the disturbance including (1) the auricular rate, (2) the configuration of the P′ wave, (3) the position of the ectopic focus, (4) the configuration of the Ta wave if one is present, (5) the presence or absence of auriculoventricular block and (6) the presence or absence of ventricular aberration. Since an abrupt change in terminology would aggravate rather than mitigate prevailing confusion, the designation auricular flutter might

be retained in instances which fulfill all the commonly accepted criteria for the diagnosis of the arrhythmia. In borderline cases a descriptive diagnosis such as that outlined provides a picture of the actual events in the auricles instead of an ambiguous term. As a fuller understanding of the nature of tachycardia and flutter is achieved it is hoped that the unrealistic verbal distinction between these arrhythmias will gradually disappear from the nomenclature and be replaced by precise descriptions of the physiologic disturbance.

EFFECT OF QUINIDINE IN AURICULAR FLUTTER

Both experimentally and clinically, quinidine has proven capable of terminating all auricular arrhythmias. Proponents of the circus movement theory have attributed the effect of quinidine on auricular flutter and fibrillation to the supposed ability of the drug to interrupt an excitable gap on the hypothetical circus pathway and thereby to restore normal rhythm.[30,31] Since the circus movement theory of flutter is invalid, a new explanation for the anti-arrhythmic action of quinidine must be sought. The basis for such an explanation lies in the concept that auricular tachycardia and auricular flutter both represent the discharge of impulses from an ectopic focus at a rate between the rate of discharge from the sinus node and the fibrillation threshold.

The effect of quinidine on auricular excitability, auricular conductivity and auricular rate was studied by means of high-speed cinematographs and/or direct and limb lead electrocardiograms recorded during quinidinization. As observed by previous workers, quinidine raises the threshold of auricular excitability at any given auricular rate.[32] When the rate of discharge from an ectopic focus is maintained at a constant level, quinidine causes a slowing of the rate at which contraction and excitation waves are conducted across the auricles. When the rate of discharge from the ectopic focus is not controlled, quinidine causes a slowing of the auricular rate which culminates in the restoration of normal sinus rhythm. These observations indicate that the primary anti-arrhythmic action of quinidine consists of a depression of auricular activity in or immediately around the ectopic focus. If the quinidine-induced slowing of the rate of discharge from the focus occurs abruptly, direct conversion to normal sinus rhythm is achieved. If the rate declines slowly, the characteristics of flutter are gradually replaced by the pattern of tachycardia, normal rhythm is restored when the ectopic focus becomes less active than the sinus node. Until more is known concerning the electrobiochemical reactions of cardiac muscle the manner in which quinidine slows the rate of discharge from ectopic foci must remain a subject of speculation. The depressing effect of quinidine on auricular excitability may cause a slowing of impulse formation in the focus and/or the conduction-depressing effect of the drug may retard the conduction of impulses out of the focus. Whatever the precise mechanism involved, it is apparent that the primary anti-arrhythmic action of quinidine occurs at the ectopic focus and not at an excitable gap on a circus pathway.

SUMMARY AND CONCLUSIONS

The results of a study of the mechanism of auricular flutter in man and experimental animal are described. By extensive application of direct and sensitive investigative techniques, the invalidity of the circus movement theory was conclusively established and the entire course of the flutter wave was demonstrated.

In the experimental animal, all portions of both auricles were exposed and auricular flutter was produced by local application of aconitine or electrical stimulation. By means of high-speed cinematographs the motion throughout the fluttering auricles was clearly visualized for the first time. Direct lead electrocardiograms from all regions of the auricles, including the hitherto unexplored body of the left auricle, revealed the entire course of the flutter excitation wave. Both the contraction and the excitation waves of auricular flutter arise from the ectopic focus and spread outward through the auricles in all available directions simultaneously. No circus movement is present. The course of the contraction and excitation waves of auricular flutter is similar to the course of the waves of auricular premature systole and auricular paroxysmal tachycardia when the arrhythmias arise from the same site.

In man, the course of the excitation wave of spontaneous auricular flutter was traced by means of esophageal and precordial lead electrocardiograms. As in the experimental animal the excitation waves of clinical auricular flutter arise at the ectopic focus and spread outward through both auricles simultaneously. No circus move-

ment is present. The speed of the excitation wave in the human auricle is slower during auricular flutter than during normal sinus rhythm.

The auricular deflection in limb lead electrocardiograms of auricular flutter consists of two components: (1) a segment which represents the entire period of auricular depolarization and is a counterpart of the P′ wave of auricular tachycardia and (2) an oppositely directed wave which represents auricular repolarization and is rendered visible by the rapidity of the auricular rate and the presence of auriculoventricular block. The first of these components is termed the P′ wave of flutter; the second is the Ta wave. A brief isoelectric period usually follows the Ta wave.

The majority of instances of clinical flutter arise in the caudal region of the auricles from whence the excitation waves travel through both auricles in a caudocephalic direction. In a minority of instances of clinical auricular flutter the excitation waves originate in the cephalic region and pursue a cephalocaudal course through both auricles. Clinical tachycardia, conversely, appears to arise most frequently in the cephalic region of the auricles and seldom originates in the caudal region.

The various electrocardiographic and therapeutic differences between auricular tachycardia and auricular flutter are related to differences in auricular rate and do not indicate a fundamental difference in mechanism of the two arrhythmias. Tachycardia and flutter represent the same fundamental disturbance in the auricles, namely, the discharge of impulses from an ectopic focus at a rate greater than the rate of discharge from the sinus node and lower than the fibrillation threshold. Within this rate range the characteristics of tachycardia tend to appear at slower auricular rates while those of flutter occur most frequently at more rapid rates. In man and experimental animal no sharp line of demarcation exists between tachycardia and flutter.

Current criteria for differentiating auricular tachycardia from auricular flutter are not applicable in borderline cases. To avoid arbitrary diagnoses in such instances a descriptive type of diagnosis is proposed which specifies the physiologic characteristics of the disturbance in the auricles.

Quinidine converts both auricular flutter and auricular tachycardia to normal sinus rhythm by slowing the rate of discharge from the ectopic focus. The concept that quinidine alters an excitable gap of a circus movement is erroneous.

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Use of Pronestyl in the Treatment of Ectopic Rhythms*

Treatment of Ninety-eight Episodes in Seventy-eight Patients

KEEHN BERRY, M.D., E. LEE GARLETT, M.D.,
SAMUEL BELLET, M.D. *and* WILLIAM I. GEFTER, M.D.
Philadelphia, Pennsylvania

A NUMBER of agents have proven useful in the treatment of supraventricular arrhythmias.[1–20] The therapy of ventricular ectopic rhythms, however, has been less satisfactory. Quinidine is acknowledged to be the most effective drug used in the treatment of ventricular ectopic rhythms[21–24] although potassium,[18,18a] magnesium,[19] morphine,[25] atropine[26] and other drugs have been tried with varying degrees of success. A great deal of attention has been paid recently to the use of procaine and its derivative, diethylaminoethanol, in the treatment of rapid ectopic cardiac rhythms.[29–36] Procaine, however, has several disadvantages: It is rapidly hydrolyzed in the body so that it is difficult to maintain a therapeutic level; it produces central nervous system stimulation in the unanesthetized individual; and it is not effective when given by the oral route. Diethylaminoethanol, a hydrolysate of procaine, although less active is safer than procaine in a corresponding effective dose. Although of rare occurrence, convulsive disorders have followed the use of diethylaminoethanol and relatively large doses must be given.

More recently procaine amide (pronestyl) has been found to be effective in the treatment of ectopic cardiac rhythms.[37] It differs structurally from procaine in that the para-aminobenzoic acid and the diethylaminoethanol are bound through an NH group rather than an ester linkage. (Fig. 1.) Pronestyl is not affected by the procaine esterase of the body and consequently is effective by either the oral or the intravenous route. Being a more stable compound than either procaine or diethylaminoethanol it has been found to give a more prolonged period of protection against cyclopropane-epinephrine induced arrhythmias in the dog.[37] Serum pronestyl levels decrease only 10 to 15 per cent per hour following a single intravenous injection and roughly 60 per cent of the drug is excreted

PROCAINE

PRONESTYL

FIG. 1. See text.

unchanged in the urine.[38] Indirect evidence indicates that it acts by depressing the myocardium as does quinidine. An antivagal effect of pronestyl has been noted in the experimental animal.[42]

The purpose of this paper is to evaluate the efficacy, optimal dosage, toxic manifestations and indications for pronestyl, and to present our experience with the drug in the therapy of various ectopic rhythms.

METHOD

Pronestyl has been used to treat ninety-eight episodes of electrocardiographically proven arrhythmias in seventy-eight patients. (Table I.) In addition one patient with a sinus rhythm who was given pronestyl intravenously because of an

* From the Division of Cardiology, Philadelphia General Hospital, Women's Medical College and Robinette Foundation, University of Pennsylvania, Philadelphia, Pa. This work was aided by a grant from the U. S. Public Health Service and E. R. Squibb & Sons.

TABLE I

CLINICAL DATA OF PATIENTS TREATED WITH PRONESTYL

Case No.*	Sex and Age	Diagnosis	Dose	Results	Remarks
			Ventricular Tachycardia		
1 (2)	M 52	Acute myocardial infarction; digitalis intoxication	1 gm. i.v. twice in 1 hr.	Failed to convert	Developed hypotension; convulsions, QRS widening; died 45 min. after 2nd injection
2 (3)	F 55	ASHD, rheumatoid arthritis	1 1/2 gm. orally q.4.h. for 2 days at a time intermittently; 1 gm. i.v.	Incomplete control VES and runs of tachycardia persist in 3 separate episodes	Hallucinations, nausea, vomiting; QRS widening
3 (1)	M 80	ASHD, diabetic acidosis, right heart failure	300 mg. i.v.; 500 mg. orally for 6 doses	NSR	Patient in shock; blood pressure returned to normal with NSR; QRS widening
4 (1)	M 59	Subacute myocardial infarct, failure	500 mg. orally q.4.h. for 5 doses	NSR	Patient in shock; blood pressure rose to 100/80 with NSR; QRS widening
5 (1)	F 64	Acute myocardial infarct, failure	150 mg. i.v.; 500 mg. orally q.4.h. for 6 doses	NSR	Posterior infarct; blood pressure rose, patient became better clinically
6 (1)	F 70	HCVD, digitalis intoxication, failure	500 mg. orally q.i.d. for 1 wk.	NSR	VES reappeared when pronestyl stopped
7 (1)	M 66	HCVD, diabetes, bidirectional ventricular tachycardia, digitalis intoxication	1 gm. i.v. in 5 min.	Restored to auricular fibrillation; no VES	No symptoms during injection
8 (2)	M 46	ASHD	1 gm. i.v. in 5 min.	Failed to convert	No toxic symptoms; QRS widening
9 (1)	M 35	Normal heart clinically	1 gm. i.v.	NSR	No toxicity
10 (1)	M 76	ASHD, digitalis intoxication	500 mg. i.v.	Stopped tachycardia	Patient's control tracing showed auricular fibrillation; following R_x for tachycardia auricles were still fibrillating
11 (1)	F 44	RHD, digitalis intoxication	1 gm. orally stat., followed by 1 gm. q.4.h. then q.3.h.	NSR	Patient had multiple multifocal VES which were partially abolished by pronestyl q.4.h.; however, 3 hr. after 4th dose ventricular tachycardia developed; dosage schedule moved to q.3.h. with complete control
12 (2)	M 100	ASHD	130 mg. i.v.; 1 gm. orally q.4.h. for 24 hr.	NSR	No toxicity
13 (2)	M 58	ASHD	1 gm. i.v.; 0.5 gm. q.4.h. for maintenance	NSR	Urticaria
14 (1)	M 17	Congenital heart	0.5 gm i.v.	Ventricular fibrillation	Patient died
			Ventricular Premature Contractions		
15-49 (35)		Ventricular premature contractions	Ranged from 250 mg. q.i.d. to 1 gm. q.4.h. from 1 day to 1 wk.	Abolished in 30, failed to abolish VES in 5 patients	1 episode of coronary insufficiency with hypotension; 2 episodes of nausea and vomiting
			Nodal Tachycardia		
50 (3)	M 57	ASHD, pulmonary emphysema	850 mg i.v., 1 gm orally stopped tachycardia; 1 gm. q.4.h. given for maintenance	NSR	Tachycardia recurred when taken off pronestyl, 3 times nausea and vomiting, urticaria
51 (3)	M 64	ASHD	400 mg. i.v.; 750 mg orally q.4 h. for maintenance	NSR	Patient returned to tachycardia when taken off pronestyl
52 (1)	M 66	ASHD, digitalis intoxication, failure	500 mg. orally q.4.h. for 5 doses	NSR	
53 (3)	F 75	HCVD	500 mg. i.v. on 3 occasions	NSR	Nodal tachycardia stopped with as little as 150 mg.
			Paroxysmal Auricular Tachycardia		
54 (1)	F 78	ASHD	400 mg. i.v.	NSR	Not stopped by carotid sinus or eyeball pressure
55 (1)	M 60	Ca. of lung with metastases to pericardium	1 gm. i.v.	No change	2:1 A-V block before and after R_x

*No. of episodes are noted in parentheses.
ASHD=arteriosclerotic heart disease.
HCVD=hypertensive cardiovascular disease.
RHD=rheumatic heart disease.
VES=ventricular extrasystoles.
NSR=normal sinus rhythm.

TABLE I (Continued)

Case No.*	Sex and Age	Diagnosis	Dose	Results	Remarks
56 (1)	F 88	Nephrosclerosis, ASHD, uremia	1 gm. i.v.	No change	2:1 A-V block before and after Rx
57 (1)	M 66	ASHD	1 gm. i.v.	No change	QRS widening with PR prolongation
58 (2)	F 45	Thyrotoxicosis	450 mg. i.v.; 250 mg. orally q.4.h. for 12 doses	NSR	Nausea, vomiting and mental depression on oral medication
59 (1)	M ?	ASHD	500 mg. converted to NSR orally q.4.h. for maintenance	NSR	None
60 (2)	F ?	ASHD	500 mg. converted to NSR orally q.4.h. for maintenance	NSR on one occasion, failed to convert on second trial	None
61 (1)	F 34	Collagen disease	150 mg. i.v.	NSR	Patient still had a sinus tachycardia of 150 after Rx
62 (1)	M 82	Silicosis	400 mg. i.v.	NSR	Hypotension; patient convulsed on 400 mg. and died 15 min. later
63 (1)	M 77	ASHD, uremia	150 mg. i.v.	NSR	None
			Auricular Flutter		
64 (1)	M 74	ASHD	1 gm. i.v.	Failed to convert	Relief of herpes zoster pain
65 (2)	F 22	RHD, active rheumatic fever	500 mg. orally q.4.h. on 2 occasions	Failed to convert	Fibrillation to flutter; flutter failed to convert
66 (4)	M 44	RHD with failure	1 gm. i.v; repeated oral doses to 1 gm. q.4.h. for 3 days	Prolonged f-f interval	Patient failed to convert after digitalis and after quinidine, 1 gm., every 2 hr.
67 (2)	M 47	HCVD with failure	1 gm. i.v.; 1.5 gm. i.v.	Prolonged f-f interval; QRS widening	Blood pressure fell from 240/110 to 80/?; patient complained of severe precordial pain; symptoms and hypotension lasted 20 min.; no sequelae
68 (1)	M 44	RHD?	1 gm. i.v.	QRS widening; prolonged f-f interval	No symptoms
69 (1)	M 55	ASHD	1 gm. i.v.	Flutter rate slowed; QRS widening; 1:1 flutter	No symptoms
70 (1)	F 60	ASHD	400 mg. i.v.	Flutter rate slowed; QRS widening; 1:1 flutter	Palpitation with 1:1 flutter
71 (1)	F 70	ASHD	500 mg. i.v.	NSR	Long P-R interval following recovery
			Auricular Fibrillation		
72 (1)	M 66	ASHD, digitalis intoxication	500 mg. orally q.4.h. 2 days	No change	None
73 (1)	M 78	Digitalis intoxication	1 gm. orally	No change	VES returned when pronestyl stopped
74 (2)	F 22	Rheumatic fever, digitalis intoxication	500 mg. orally q.4.h. for 2 days on 2 occasions	Auricular flutter	Psychosis; back to fibrillation when off pronestyl
75 (1)	M 66	HCVD, diabetes	1 gm. i.v.	No change	None
76 (1)	F 44	RHD, digitalis intoxication	1 gm. stat. i.v.; 500 mg. q.4.h. orally for 2 days	No change in auricular mechanism	
77 (1)	M 76	ASHD	500 mg. i.v.	No change in auricular mechanism	
78 (1)	M ?	ASHD and paroxysmal auricular fibrillation	300 mg. i.v.; 500 mg. orally for 1 dose	NSR but patient continued to fibrillate intermittently	None
			Sinus Tachycardia with Widened QRS Complexes Mistaken for Ventricular Tachycardia		
79 (1)	M 60	ASHD, HCVD, acute myocardial infarct	400 mg. i.v.	Cardiac arrest	

*No. of episodes are noted in parentheses.
ASHD=arteriosclerotic heart disease.
HCVD=hypertensive cardiovascular disease.
RHD=rheumatic heart disease.
VES=ventricular extrasystoles.
NSR=normal sinus rhythm.

error in diagnosis is included in Table I. The method of administration was as follows: The drug was administered intravenously twenty-four times (to twenty-one patients); intravenously until the ectopic rhythm was abolished, followed by oral maintenance twenty-six times (to twelve patients); and by the oral route alone forty-eight times (to forty-five patients). The intravenous preparation was administered at a rate of not more than 200 mg. per minute under constant electrocardiographic control until the electrocardiogram showed return to normal sinus rhythm, until toxic manifestations (to be discussed later) were observed or until a total of 1 gm. had been given. In one patient (Case 67) a total dose of more than 1 gm. was injected by vein, and in another patient (Case 1) a second 1 gm. dose was given one hour after an initial 1 gm. intravenous dose. Frequent blood pressure readings were made during the period of administration.

The effective oral dose varied widely (Table I) and was established largely by trial. The usual dose given was 0.5 to 1 gm. every four hours. With the exception of one patient (Case 2) who received 1.5 gm. every four hours and another (Case 11) who received 1 gm. every three hours, the maximum dose was 1 gm. every four hours. Indications for stopping oral medication were clinical or electrocardiographic evidence of toxicity, or failure to convert the arrhythmia to a normal sinus rhythm within twenty-four hours. Many of the patients who were converted to a normal sinus rhythm by either oral or intravenous administration of the drug were maintained for varying lengths of time on oral medication to prevent recurrence of the arrhythmia.

Because rapid ectopic rhythms are capable in themselves of altering the electrocardiogram after conversion to a sinus rhythm, the electrocardiographic effects of pronestyl were determined in twenty-seven cases (twenty-one patients received the drug orally and six intravenously) with a normal sinus rhythm.

RESULTS

The results of therapy in the various types of ectopic rhythm are shown in Table II.

Ventricular Tachycardia. In all but three episodes of ventricular tachycardia pronestyl was administered by the intravenous route. In eight of the eleven patients (seventeen episodes) who received pronestyl parenterally normal sinus rhythm was restored during the injection and recorded by the electrocardiogram. Figures 2 and 3 show the electrocardiograms of successfully treated patients. Digitalis intoxication was probably implicated in five of the patients (six episodes) with ventricular tachycardia; in one of these (Case 7) bidirectional tachycardia was present. Pronestyl was effective in restoring normal rhythm in four of the six episodes. Three of the episodes in a single patient (Case 2, Table I) with ventricular tachycardia are labeled "partial control." In these incompletely controlled patients pronestyl was successful in abolishing the arrhythmia but maintenance doses as high as 1.5 gm. every four hours did not prevent recurrence.

TABLE II

EFFECT OF PRONESTYL IN THERAPY OF ARRHYTHMIAS IN SEVENTY-EIGHT CASES (NINETY-EIGHT EPISODES)

Arrhythmias	Cases	Episodes	Return to Sinus Rhythm*	Partial Control	Failed
Ventricular tachycardia	14	20	12	3	5
Multiple premature ventricular contractions	35	35	30		5
Nodal tachycardia	4	10	10†		0
Paroxysmal auricular tachycardia	10	12	8		4
Auricular flutter	8	13	1		12
Auricular fibrillation	7	8	0		8
Total	78	98	61		34

* In two of these in which the ventricular tachycardia was associated with auricular fibrillation the rhythm was restored to auricular fibrillation.

† Recently we have had two failures.

Ventricular Premature Contractions. Pronestyl was successful in abolishing the arrhythmia in thirty of thirty-five episodes (in thirty of thirty-five patients) with multiple ventricular premature contractions. In most of these patients the premature contractions returned when the drug was stopped.

Supraventricular Arrhythmias. Pronestyl has proven effective in the treatment of paroxysmal auricular and nodal tachycardias. (Fig. 4.) There were eighteen successes and four failures in these two groups. (Table II.) Only one of thirteen episodes of established auricular flutter and none of eight episodes of chronic auricular fibrillation could be converted to a normal sinus rhythm. Pronestyl resulted in a slowing of the flutter rate with an increase in the ventricular rate. This series of events resulted in a 1:1 A-V conduction in two patients. Figure 5 shows the

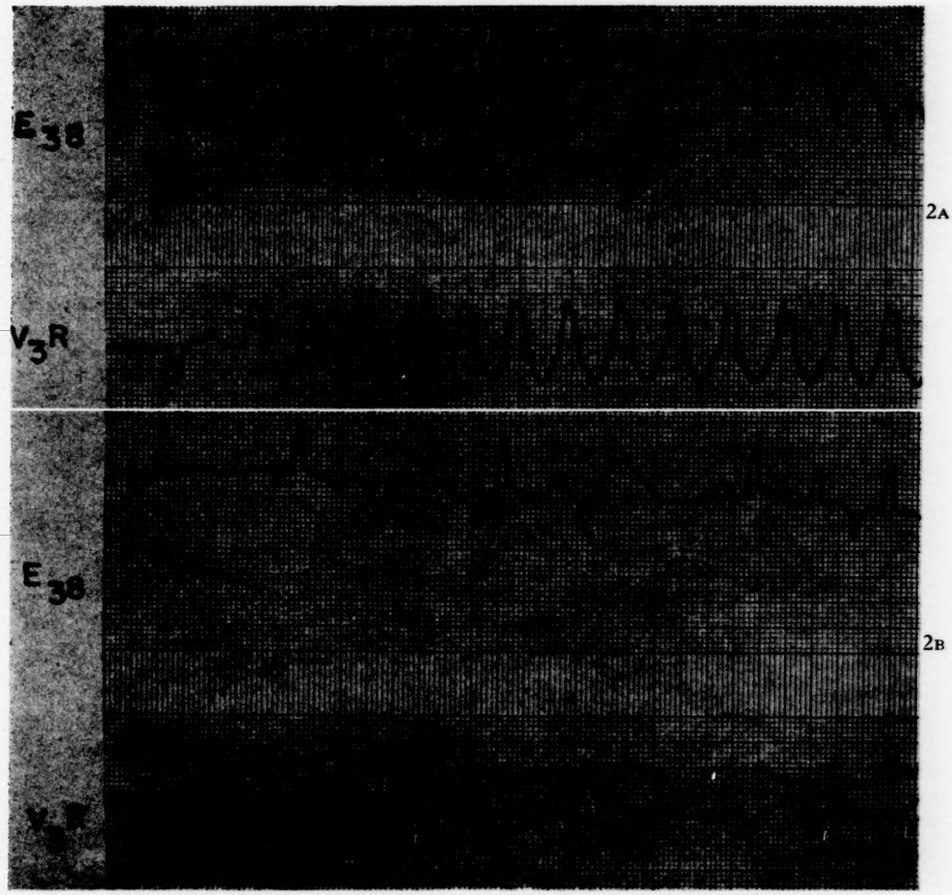

FIG. 2. Case 12. Simultaneous esophageal (E_{38}) and precordial (V_3R) leads. A, shows ventricular tachycardia (control). Note that the esophageal lead reveals retrograde conduction with progressive prolongation of the R-P interval until a retrograde P is dropped. B, taken during the administration of intravenous pronestyl, shows change to a bigeminal rhythm and subsequent establishment of a sinus rhythm which persisted after pronestyl.

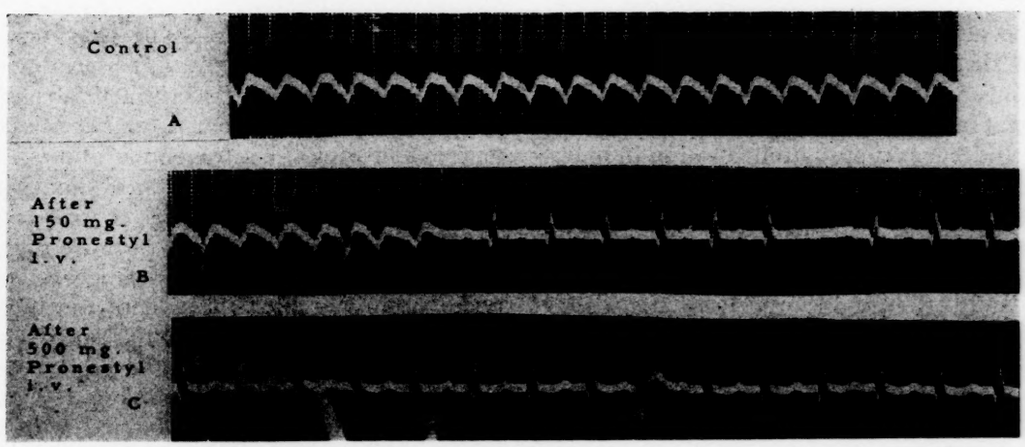

FIG. 3. Case 5. Ventricular tachycardia showing conversion to a sinus rhythm after the administration of 150 mg. of pronestyl intravenously (continuous tracing).

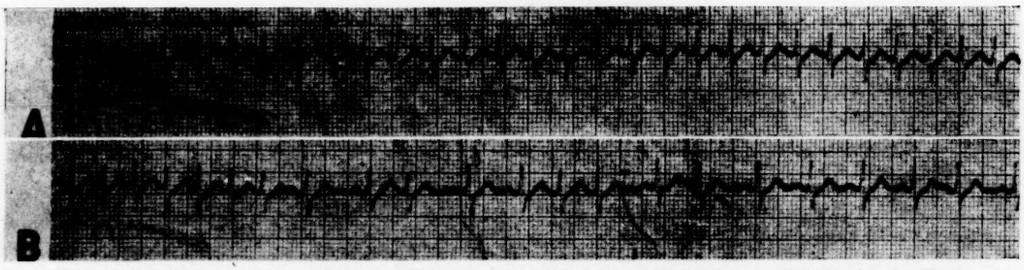

FIG. 4. Case 58. Paroxysmal auricular tachycardia. A, control; B, after pronestyl (400 mg. in four and a half minutes); shows conversion to a sinus rhythm with occasional auricular premature contractions after 400 mg. of pronestyl intravenously.

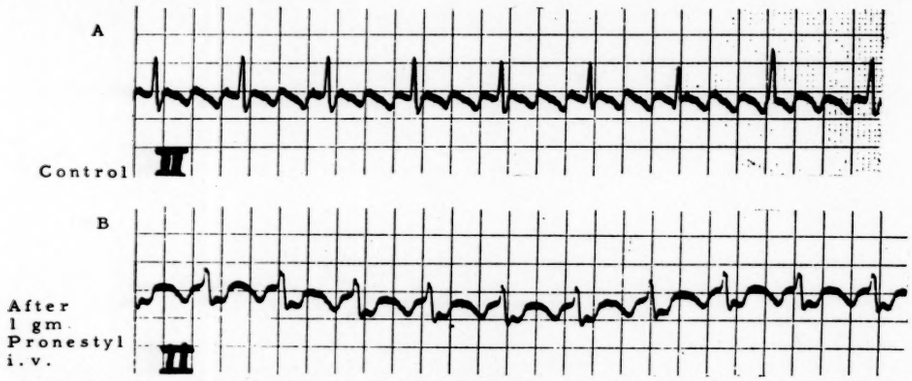

FIG. 5. Case 67. A, Auricular flutter with 3 :1 block; B, shows slowing of the flutter rate and an increase in the ventricular rate after pronestyl.

usual response in a patient with auricular flutter. This effect is similar to that seen with quinidine although generally the auricular rate is slowed more by therapeutic doses of quinidine than it is by pronestyl.

Toxic Reactions. In the seventy-eight patients who received pronestyl in the treatment of 98 episodes of ectopic cardiac rhythms there were fifteen episodes of toxicity; in addition there were four patients who were in extremis in whom the administration of pronestyl was followed by death within forty-five minutes after the administration of the drug. Oral administration was associated with toxicity in eleven episodes and intravenous administration in four. These figures cannot be taken to indicate that the oral is more toxic than the intravenous preparation since the majority of the patients in the series, fifty-seven, received oral medication whereas only twenty-one received the drug intravenously. Intravenous medication consisted in all but two instances of a single injection in any twenty-four-hour period whereas the oral preparation was administered over a period of time varying from twenty-four hours to several weeks. In general the toxic manifestations following intravenous administration of the drug were more alarming when they appeared than were those in patients given the oral preparation.

Fall in blood pressure was the most serious untoward manifestation, and practically all patients who received the drug by vein manifested some degree of hypotension. A significant drop of blood pressure occurred occasionally after the injection of as little as 100 mg. Since many of these patients are already in some degree of shock, a further reduction in blood pressure could have serious consequences. The hypotension was accompanied with alarming reactions in four patients. Severe anginal pain was produced in one patient who became hypotensive following the intravenous administration of the drug. This patient (Case 67) experienced a fall in blood pressure from control levels of 240/110 to 80/?. The blood pressure started to rise and the precordial pain disappeared within twenty minutes after the end of the injection. In one patient (Case 1) convulsions developed affecting the extremities involved by an old hemiparesis, and in another patient (Case 62) generalized convulsions developed with a fall in blood pressure. Decreasing the rate of intravenous administration and having the patient lie flat during and for a time after the injection

diminished but did not entirely abolish the hypotensive effect. No definite correlation could be made between the size of the dose and the degree of fall in blood pressure although, in general, larger doses produced a more marked effect. This hypotensive effect is most pronounced in patients with hypertension and those with severe myocardial disease. In discussing the blood pressure changes observed as the result of pronestyl administration it should be remembered that in instances in which hypotension or a shock-like state are secondary to rapid arrhythmia, conversion to a normal sinus rhythm, whether spontaneous or drug induced, will result in return of the blood pressure toward normal.

The reduction in blood pressure was uncommonly seen in patients given the oral preparation although in one patient who was being treated for ventricular premature contractions hypotension developed and there was electrocardiographic evidence of extension of an old myocardial infarction coincident with the fall in blood pressure while on oral pronestyl for the treatment of multiple ventricular premature contractions. The hypotension, change in the electrocardiogram, nausea, vomiting and postural syncope disappeared within twenty-four hours after the drug was stopped.

Two patients (Cases 2 and 65) developed definite psychoses and a third (Case 58) severe mental depression while on pronestyl. Mental aberrations in the form of visual and auditory hallucinations and paranoid ideas were reproduced repeatedly in the patients in whom psychosis appeared. These symptoms subsided completely within twenty-four hours of stopping therapy. Severe mental depression developed in the third patient (Case 58) while she was receiving 250 mg. of the oral preparation every four hours to prevent recurrent episodes of paroxysmal auricular tachycardia. It is interesting to note that each of these three patients had an active disease process at the time of treatment. One had active rheumatic fever, another had active rheumatoid arthritis and the third was hyperthyroid.

Nausea and vomiting developed in six episodes which were being treated by oral administration of the drug. This figure is not necessarily an accurate reflection of the incidence of nausea and vomiting due to pronestyl because many of the patients treated were experiencing these symptoms due to other causes—digitalis intoxication, uremia before therapy with pronestyl was begun. In these six episodes, however, it was believed that pronestyl could be definitely implicated as the cause of the untoward symptoms. Therapy was stopped with the appearance of nausea and vomiting in these patients. At present we believe that these symptoms *per se* do not necessarily warrant stopping the administration of the drug.

Urticaria developed in two patients, one on oral medication and the other following intravenous administration.

Four of the patients in this series died during or within forty-five minutes of intravenous administration of pronestyl. These patients were in extremis when treated and it is difficult to ascertain whether or not the drug might be directly implicated in the death of the patient. Because this drug is relatively new and is being used widely, these cases merit some discussion.

Case 1. A fifty-two year old male was admitted with severe digitalis intoxication attributable to taking 0.8 mg. of digitoxin per day for the fourteen days prior to admission. On the morning of admission the patient experienced severe substernal pain and collapsed. Significant findings on admission included marked congestive heart failure, a markedly enlarged heart and an electrocardiogram showing ventricular tachycardia and suggestive of an acute posterior wall myocardial infarction. One gram of pronestyl was given intravenously without demonstrable effect. Approximately one hour after the first dose another gram was given. The second dose also failed to convert the arrhythmia. Because of marked QRS widening it was not believed advisable to superimpose the effects of quinidine on those of pronestyl. Since this tachycardia could conceivably have been secondary to digitalis intoxication, for which potassium has been recommended, 500 cc. of isotonic potassium chloride were given by vein in forty-five minutes. While the potassium chloride was being given, approximately forty minutes after the second injection of pronestyl, ventricular fibrillation developed and the patient died. Postmortem examination showed a large, fresh posterior myocardial infarct.

Case 79. This patient, a sixty year old white male, gave a history of hypertension and a "heart attack" four years before the present admission. He was admitted with a two-day history of severe persistent precordial pain, and an electrocardiogram showed a left bundle

branch block. Ten days after admission he suddenly complained of the onset of severe, crushing substernal pain which radiated down both arms. He became cyanotic, disoriented, cold and clammy with a mottled skin, and the blood pressure was unobtainable. The electrocardiogram at this time showed a sinus rhythm with widened QRS complexes but was misinterpreted as showing ventricular tachycardia and the patient was given 400 mg. of pronestyl by vein over a five-minute period. After 400 mg. had been injected and while the needle was still in the vein, the patient went into asystole and expired. Postmortem examination showed hypertrophy of both ventricles (the heart weighed 760 gm.). The coronary vessels were markedly sclerotic; the orifice of the anterior descending branch of the left coronary artery failed to permit the entrance of a small probe. There was marked diffuse fibrosis and a fresh infarction involving the anterior wall of the left ventricle and adjacent portion of the interventricular septum.

CASE 62. This patient, an eighty-two year old white male, was admitted in a state of shock. No history was available but a routine chest plate on admission showed far advanced silicosis and tuberculosis. Paroxysmal auricular tachycardia was noted on the admission electrocardiogram. This tachycardia was not affected by carotid sinus pressure; and since it was thought that immediate restoration of sinus rhythm was necessary, 400 mg. of pronestyl were given intravenously over a five-minute period. With the restoration of sinus rhythm the patient was seized with a generalized convulsion. Fifteen minutes later respiratory arrest was noted. A normal rhythm persisted for five minutes after the respiratory arrest appeared. The terminal cardiac mechanism was cardiac standstill. Permission for postmortem examination was not obtained.

CASE 14. A 17 year old patient with cyanotic form of congenital heart lesion (tetralogy of Fallot?) was admitted to the Accident Ward in extremis. Because of the presence of ventricular tachycardia an initial dose of pronestyl was given by vein (300 mg. in five minutes). This dose of pronestyl was followed with still further widening of the ventricular complexes which became more bizarre. Two hundred milligrams more were administered by vein, but widening progressed until finally fibrillation ensued. Because of the critical state of the patient and the type of arrhythmia, it was believed that pronestyl was only partially responsible for the outcome in this case.

Electrocardiographic Effects. In patients with an ectopic rhythm who received pronestyl electrocardiographic changes other than conversion of the abnormal rhythm included slowing of the ventricular rate in ventricular tachycardia and slowing of the flutter rate in auricular flutter. Other electrocardiographic changes were observed in twenty-seven patients with a sinus rhythm who were given oral or intravenous doses. In general the effects following pronestyl are the same that others have noted with the use of procaine[39] or quinidine[40] and consist of PR and QT prolongation, T wave flattening and QRS widening. Such changes were usually observed following intravenous administration of the drug and although infrequent are seen often enough with oral medication to warrant electrocardiographic control. It is our impression that these alterations are apt to be more pronounced in patients with extensive myocardial disease and in patients who have been receiving the drug for more than twenty-four hours.

QRS widening of considerable degree with either intravenous or oral medication is an indication for stopping therapy.

Peak effect after intravenous administration as measured by increase in QTc[43] is noted four minutes after the end of injection. Following oral medication it is noted within one and two hours.

COMMENTS

Qualifications of an Antifibrillatory Drug. An ideal drug for the treatment of rapid ectopic rhythms should prolong the effective refractory period of the myocardium, should not produce areas of local block and should abolish this latter abnormality if present, and should not slow conduction through the specialized tissues of the heart.[41] In addition it should be a rapidly acting compound that is non-toxic in effective therapeutic doses. Pronestyl, then, cannot be considered the perfect agent for the treatment of cardiac arrhythmias. The electrocardiographic changes previously described indicate that a slowing of conduction may occur in the auricle, the auriculoventricular node and its ramifications, and in the ventricular myocardium. It may produce untoward side effects in therapeutic doses and has not been effective in all of the arrhythmias treated. These disadvantages,

however, while seemingly great are characteristic of other preparations, particularly quinidine, procaine and diethylaminoethanol, that have been extensively used in the treatment of arrhythmias.

Place of Pronestyl in Therapy of Ectopic Rhythms. Although pronestyl has proven effective in the therapy of paroxysmal auricular and nodal tachycardias, these arrhythmias will often respond to a variety of other measures.

Pronestyl has been notably unsuccessful in the conversion of auricular flutter and fibrillation, and it is believed that its use in the therapy of these arrhythmias is not indicated. Administration of the drug to a patient with auricular flutter may produce 1:1 auriculoventricular conduction and in many patients such a rapid response might prove dangerous.

The most widely and successfully used drug in the therapy of ventricular tachycardia hitherto has been quinidine. Intravenous pronestyl has proven effective in the therapy of ventricular tachycardia and we believe that it is much less toxic than intravenous quinidine in therapeutically effective dosage. The intravenous preparation was used in the majority of the patients in this group because it was believed that this route allowed more accurate evaluation of the effects of the drug. The toxic manifestations of intravenous pronestyl are similar in many respects to those seen with quinidine although they are much less marked. Because of the rarity of serious side effects with oral pronestyl it is believed that this route is the method of choice when pronestyl is used. Because only three patients in this series with ventricular tachycardia were treated with oral medication, no accurate comparison can be made with quinidine administered by the same route. All three of these patients converted to sinus rhythm. Pronestyl is of further value in that some patients[38] who have not responded to quinidine given for the treatment of ventricular tachycardia have converted to sinus rhythm following the use of pronestyl. Pronestyl should probably supplant procaine and diethylaminoethanol in the treatment of ventricular tachycardia.

All cases of ventricular premature contractions do not require therapy but pronestyl has proven useful in the treatment of ventricular extrasystoles when therapy is deemed necessary.

Pronestyl would seem to be a logical drug to use in prophylaxis of arrhythmias following myocardial infarction but until more data are available as to the efficacy and safety of pronestyl in such a situation it would seem advisable to use the drug only when these arrhythmias appear.

Because of the severity of toxic effects observed during intravenous administration it is suggested that the rate of injection should not exceed 100 mg. per minute, and an even slower rate is recommended. Although four deaths have been reported in this series, the terminal condition of the patients at the time of treatment makes the role of pronestyl in these deaths difficult to evaluate. Preliminary observations (work now in progress) indicate that in unanesthetized dogs the mechanism of death is either by respiratory arrest or ventricular fibrillation.

The question arises as to when toxic manifestations preclude the further use of pronestyl. When conversion of the arrhythmia is an absolute necessity, minor toxic signs such as nausea, vomiting and urticaria do not warrant cessation of therapy. Obviously, such dramatic symptoms as convulsions, respiratory stimulation or depression, angina and marked QRS widening indicate immediate discontinuance of the drug. When, however, the conversion of the arrhythmia or prevention of recurrence of an arrhythmia is of less importance, untoward minor symptoms may be an indication for stopping pronestyl. Such instances are seen in patients who are being treated for paroxysmal auricular and nodal tachycardias and for premature ventricular contractions.

Pronestyl is a new and potent drug. Until more experience has been obtained with it its use should be controlled by careful electrocardiographic and clinical observations.

SUMMARY

Our experience in the treatment of ninety-eight episodes of ectopic rhythm in seventy-eight patients is discussed. Pronestyl was used in the treatment of the following arrhythmias: ventricular tachycardia, nodal tachycardia, paroxysmal auricular tachycardia, auricular flutter, auricular fibrillation and multiple premature ventricular contractions. Pronestyl was effective in converting a large proportion of these arrhythmias to normal. It was notably unsuccessful in converting auricular flutter or auricular fibrillation to a normal rhythm.

Although pronestyl is effective when administered by the intravenous route, administra-

tion by the oral route is considered to be the method of choice in treatment.

The most serious toxic manifestation is the development of hypotension which in two patients resulted in the production of coronary insufficiency and in two others produced convulsive seizures. Because of the production of hypotension frequent blood pressure determinations as well as a continuous electrocardiogram should be obtained during the intravenous administration of pronestyl. It is suggested that the intravenous preparation be given no faster than 100 mg. per minute.

Mental symptoms were observed in three patients. These patients had active toxic processes.

It is believed that pronestyl administered by vein is safer than the corresponding effective dose of quinidine administered by the same route.

Pronestyl is considered to be an important addition to the drugs available for the treatment of ectopic rhythms, particularly those of ventricular origin.

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Effect of Isuprel (Isopropylepinephrine) on Circulation of Normal Man*

JOSEPH KAUFMAN, M.D., ARNOLD IGLAUER, M.D. *and* GISELA K. HERWITZ, B.A.
Cincinnati, Ohio

RECENT reports[1,2] have suggested that isuprel® (isopropylepinephrine, aludrine) has certain advantages over epinephrine in the treatment of paroxysms of bronchial asthma. In both human[1–4] and animal[5,6] studies isuprel produced relaxation of bronchospasm without marked increase in blood pressure although increase in pulse rate often occurred. As the compound might be used in the treatment of asthma for long periods of time in patients with organic heart disease, it seemed important to know its effect on cardiac output. Since no data on the effect of isuprel on cardiac minute volume in man was available, the present study was undertaken using a high frequency, magnetically damped ballistocardiograph.[7] This instrument seemed ideally suited for study of the effect of drugs on output since continuous observations could be made with little difficulty for the observer and no discomfort to the patient.

® Trade mark.

TABLE I
PAREDRINE INTRAMUSCULAR

Patient	Age	Body Surface (M^2)	Dose (mg.)	Cardiac Output (L./min.)		Blood Pressure	
				Before Paredrine	Maximum Change after Paredrine	Before Paredrine (mm. Hg)	Maximum Change after Paredrine (mm. Hg)
S. E.	33	2.06	15	6.7	7.0	115/75	140/80
T. A.	27	1.97	15	7.0	6.9	126/70	150/74
C. N.	39	1.98	15	6.6	7.2	112/76	122/82
J. W.	40	1.76	15	7.0	7.2	130/90	158/90
C. B.	20	1.56	20	4.8	5.0	115/90	190/95

MATERIALS AND METHODS

Isuprel was administered sublingually, subcutaneously or by aerosol inhalation to normal subjects. In a study of the validity of the method paredrine (ρ-hydroxy-α-methyl-phenylethylamine hydrobromide) was administered intramuscularly to five subjects in dosage of 15 to 20 mg. and epinephrine (0.5 to 1.0 mg.) was given to four subjects. Experiments were carried out in the basal state after a minimum rest period of fifteen minutes. Records were made with a high frequency ballistocardiograph of the Starr type. Cardiac output was calculated from the curve of movement in the long axis of the body by the formula:

TABLE II
EPINEPHRINE INTRAMUSCULAR

Patient	Age	Body Surface (M^2)	Dose (mg.)	Cardiac Output (L./min.)		Blood Pressure	
				Before Epinephrine	Maximum Change after Epinephrine	Before Epinephrine (mm. Hg)	Maximum Change after Epinephrine (mm. Hg)
F. H.	20	....	1	8.1	9:3	120/70	132/65
C. N.	39	1.98	0.5	7.0	8.6	121/79	120/70
S. E.	33	2.06	0.5	7.0	8.3	114/71	148/60
J. W.	40	1.76	0.5	6.3	8.6	.....	.....

$$Vs = K\sqrt{(31 + 2J)\ C^{3/2}}$$

Vs = Stroke volume
K = Constant for particular ballistocardiograph
I = Height of I wave
J = Height of J wave
C = Duration of cardiac cycle in seconds

* From the Cardiac Laboratory, University of Cincinnati College of Medicine, Cincinnati General Hospital. Presented in part at a regional meeting of the American Federation of Clinical Research, Chicago, Ill., November 2, 1950. Aided by a grant from the National Heart Institute of the U. S. Public Health Service.

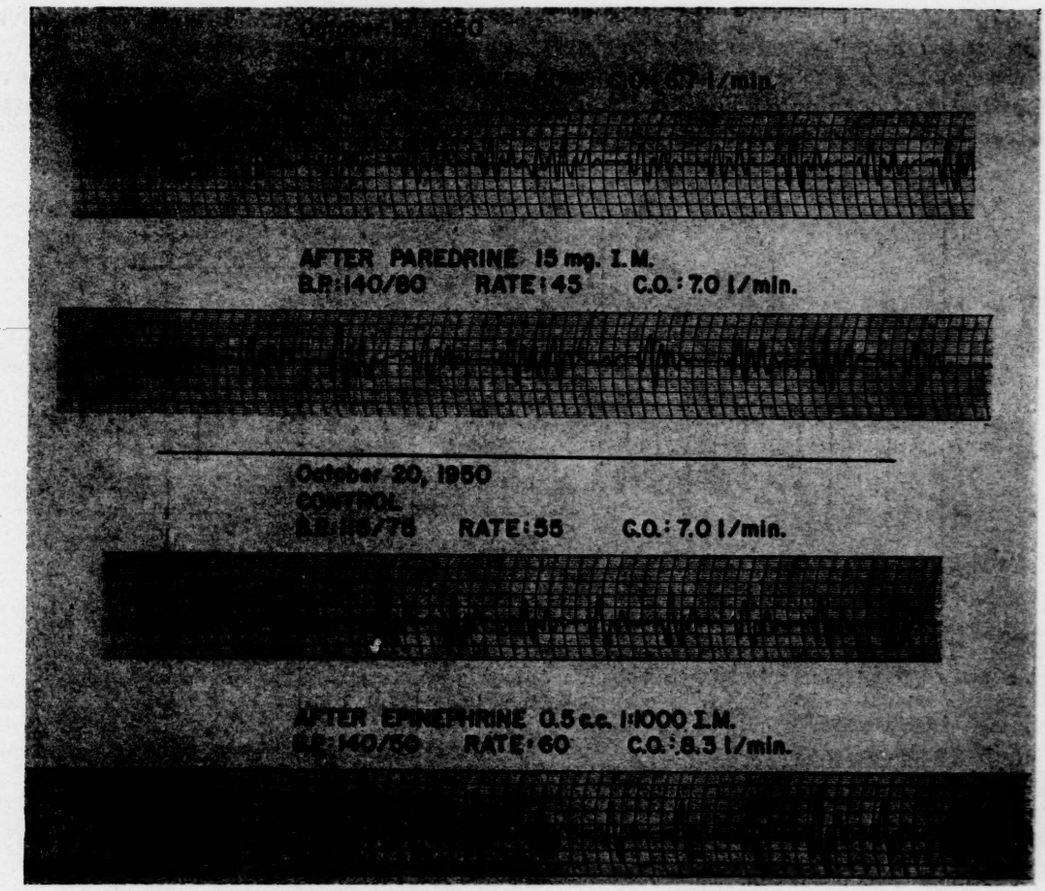


FIG. 1. Ballistocardiograms after administration of paredrine and epinephrine to same subject, J. C., age thirty-three; height of complexes unchanged by paredrine despite blood pressure increase. After 0.5 mg. of epinephrine, with same increase in blood pressure, height of complexes definitely increased.

Prior to drug administration three control records were made; following administration continuous readings were made for a minimum of two minutes and repeated at one- to three-minute intervals for thirty minutes. Changes in minute volume of more than plus or minus 10 per cent were considered significant. The aerosol was administered with a No. 40 DeVilbiss atomizer using 1:200 aqueous solution of isuprel. The solution was nebulized either by manually compressing the rubber bulb six to eight times or by a continuous stream of oxygen. Pulse rate was determined from the electrocardiogram. Blood pressure was measured every two to three minutes by the auscultatory method. Oxygen consumption was determined by means of a Benedict-Roth apparatus.

RESULTS

Paredrine, Epinephrine—Validity of Method. Paredrine in doses adequate to produce a definite increase in blood pressure caused no significant change in cardiac output as measured from the ballistocardiograph tracing. (Table I, Fig. 1.) Following administration of epinephrine, however (Table II, Fig. 1), there was a significant increase in cardiac output in all of the subjects. It was known from previous studies by another method[8] that administration of paredrine does not change cardiac output whereas epinephrine is known to increase cardiac output.[9] Thus since changes in blood pressure did not modify the cardiac output computed from the ballistocardiographic record, the suitability of the method for the present study was considered to be established.

Isuprel. Sublingual: Isuprel (15 mg.) was administered sublingually to eight different subjects. (Table III, Fig. 2.) The cardiac output increased more than 10 per cent in six subjects, was unchanged in one and decreased more than 10 per cent in one subject.

Heart rate increased more than ten beats per minute in six of the subjects; in two the rate changes were minimal. The largest increase was thirty-one beats per minute. Systolic blood pressure increased in six of the eight subjects and patients. The largest pulse pressure difference recorded was 48 mm. Hg.

No change in oxygen consumption was noted in two patients in whom determinations were obtained.

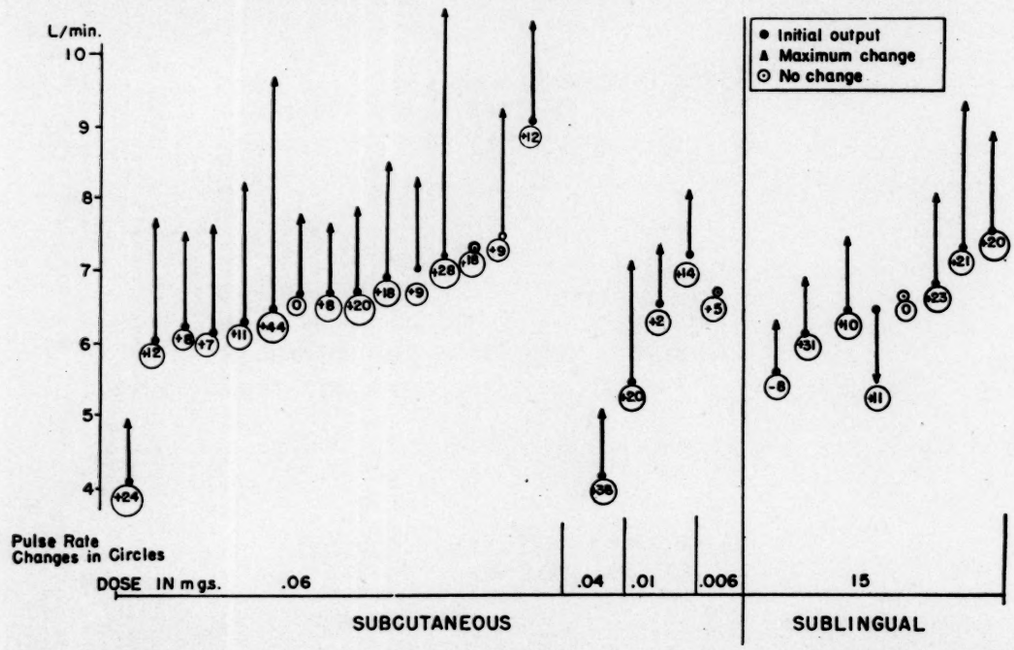


FIG. 2. Effect of isuprel on cardiac output and pulse rate. Arrowhead indicates maximal change in cardiac output after isuprel.

TABLE III
ISUPREL SUBLINGUAL

Patient	Age	Body Surface (M^2)	Dose (mg.)	Cardiac Output (L./min.)		Pulse Rate		Blood Pressure	
				Before Isuprel	Maximum Change after Isuprel	Before Isuprel	Maximum Change after Isuprel	Before Isuprel (mm. Hg)	Maximum Change after Isuprel (mm. Hg)
E. H.	35	1.78	15	6.4	+1.0	70	+10	107/77	None
R. Sc.	23	1.98	15	6.8	+1.2	53	+23	105/81	+15/−11
D. M.	24	1.71	15	6.1	+0.7	61	+31	104/60	+30/−12
O. H.	43	1.69	15	6.6	0	82	0	137/89	None
R. R.	27	2.02	15	6.4	−1.0	53	+11	110/78	+10/+2
E. G.	31	2.02	15	5.5	+0.7	76	−8	100/56	+12/+24
H. R.	13	1.58	15	7.5	+1.4	85	+20	134/80	+10/+4
E. J.	19	1.82	15	7.3	+2.0	69	+21	100/70	+18/−30

was unchanged in two. Diastolic blood pressure increased in one, fell in three and remained unchanged in four. An increase in pulse pressure of more than 20 mm. Hg was observed in four

Subcutaneous: Isuprel (.06 to .006 mg.) was administered subcutaneously to twenty patients. (Table IV, Fig. 2.) A significant rise in cardiac output occurred in eighteen subjects; the re-

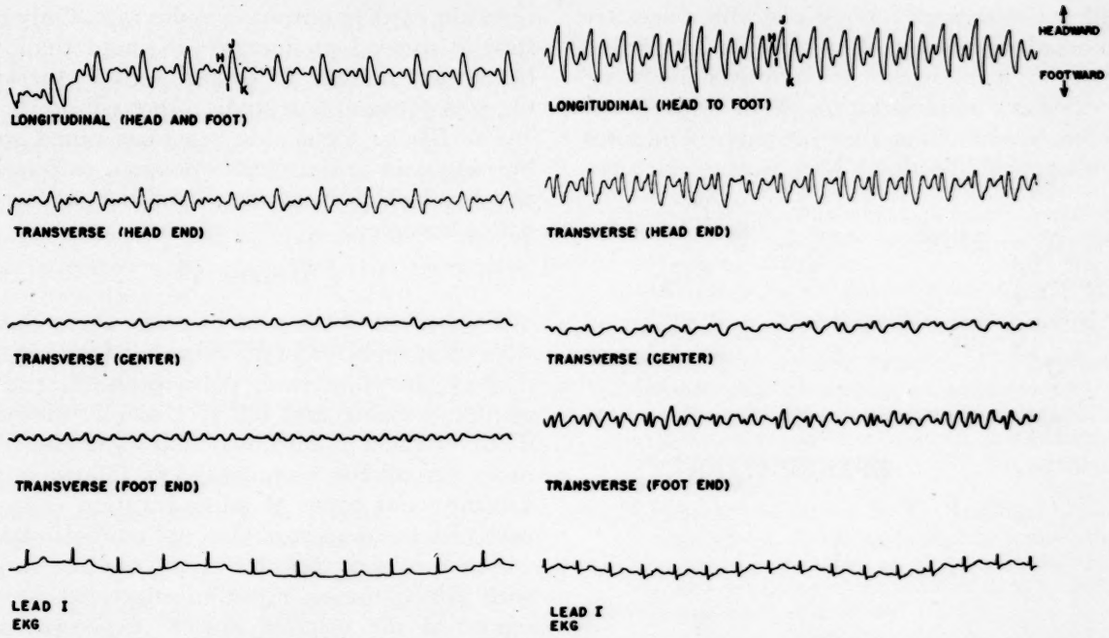


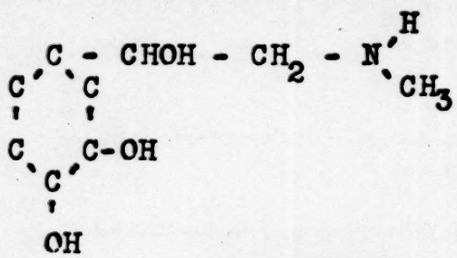
FIG. 3. Effect of subcutaneous administration of isuprel. The record shows increase not only in heart rate but also in the amplitude of movement in the long axis of the body (top tracing). There is also increased amplitude of movement in the transverse axis. A, ballistocardiogram before injection; B, ballistocardiogram five minutes after subcutaneous injection of 0.3 cc. isuprel (1:5,000).

TABLE IV

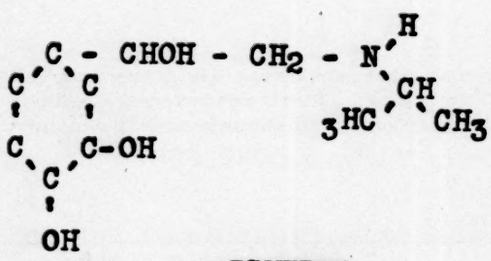
ISUPREL SUBCUTANEOUS

Patient	Age	Body Surface (M^2)	Dose (mg.)	Cardiac Output (L./min.)		Pulse Rate		Blood Pressure	
				Before Isuprel	Maximum Change after Isuprel	Before Isuprel	Maximum Change after Isuprel	Before Isuprel (mm. Hg)	Maximum Change after Isuprel (mm. Hg)
A. W.	30	1.79	.06	4.0	+0.9	76	+24	95/70	+21/−6
G. P.	28	1.72	.06	6.0	+1.7	64	+12	112/76	+36/−12
S. R.	20	2.0	.06	6.1	+1.4	52	+8	130/100	+10/−10
M. E.	25	1.81	.06	6.1	+1.5	59	+7	110/60	+20/−8
H. C.	23	1.84	.06	6.2	+2.0	94	+11	114/78	+16/−28
H. B.	23	1.43	.06	6.4	+3.2	68	+44	94/68	+22/−14
K. P.	39	1.86	.06	6.6	+1.1	75	0	119/78	+9/−16
F. D.	33	2.08	.06	6.6	+1.0	58	+8	107/73	+25/−7
L. B.	26	1.80	.06	6.6	+1.2	64	+20	123/75	+41/−15
E. D.	29	1.93	.06	6.8	+1.7	60	+18	138/89	0
G. C.	39	2.02	.06	7.0	+1.1	72	+9	107/72	+23/−10
R. K.	23	1.76	.06	7.1	+3.5	90	+28	110/72	+22/−16
S. F.	29	2.16	.06	7.3	0	51	+18	142/87	+24/−7
A. N.	27	1.79	.06	7.4	+1.8	73	+9	108/64	+16/−8
R. S.	25	2.37	.06	9.0	+1.4	68	+12	109/69	+15/−13
H. C.	23	1.84	.04	4.1	+0.9	90	+38	.....	.....
H. B.	23	1.43	.01	5.4	+1.7	68	+20	87/68	+7/−8
F. D.	33	2.08	.01	6.5	+0.8	60	+2	110/68	+2/+2
D. G.	22	1.92	.01	7.2	+0.9	64	+14	104/65	+2/−7
F. D.	33	2.08	.006	6.7	0	60	+5	115/72	+1/−6

maining two showed a slight rise which was not considered significant. The increase in cardiac output occurred within six minutes following subcutaneous administration in all subjects except one in whom it occurred twenty-six minutes following the injection. The average increase



EPINEPHRINE



ISUPREL

FIG. 4. Structural formulas of epinephrine and isuprel.

in the fifteen patients who were given .06 mg. was 26 per cent. A constant pattern in the ballistocardiogram, consisting of increased amplitude of the J and especially the K wave, was noted in all subjects who manifested a marked change in cardiac output. (Fig. 3.)

In eleven patients the heart rate increased more than ten beats per minute. The largest increase was forty-four beats per minute. The blood pressure was measured in nineteen patients. The systolic pressure increased more than 10 mm. Hg in twelve patients and the diastolic pressure fell more than 10 mm. Hg in eight patients. An increase in pulse pressure of more than 20 mm. Hg was observed in thirteen patients. The largest pulse pressure difference recorded was 56 mm. Hg. No change in oxygen consumption occurred in four patients in whom measurements were obtained.

Inhalation: Eight patients were instructed to inhale six to eight times a 1:200 strength aqueous solution of isuprel. No significant change was noted in cardiac output or pulse rate. Only one subject showed an increase of more than ten beats per minute following aerosol therapy. Oxygen consumption studies were not done.

Side Effects: Chief side reactions noted were burning pain at the site of injection, palpitation and headache. No nausea, vomiting, tremor or pallor occurred.

COMMENTS

The results of the present study agree closely with those reported in the literature[1–4] as regards increase in pulse rate, pulse pressure, rise in systolic pressure and fall in diastolic pressure. These changes in cardiovascular dynamics were more prominent and consistent following the subcutaneous route of administration than by aerosol inhalation or sublingual administration.

Our results confirm the rise in cardiac output with subcutaneous injection suggested in the report of the original animal experiments.[5,6] Our failure to obtain changes in cardiac output with aerosol administration may be related to the dosage employed and the method of administration. With relatively small doses given parenterally changes in cardiovascular dynamics indicated that like epinephrine[9] isuprel produced increased blood flow and decreased peripheral resistance.

Isuprel, like epinephrine which it resembles closely in structure (Fig. 4), produced increased cardiac output. Unlike epinephrine isuprel failed to produce blanching of the skin, tremor, increase in blood sugar or change in oxygen consumption.

Our data would suggest that like epinephrine isuprel, especially if used subcutaneously, should be employed with caution in the treatment of asthmatic patients with heart disease. The absence of changes in cardiovascular dynamics following aerosol administration confirms the clinical observations of Gay and Long[4] that this is the safest method of administration.

SUMMARY AND CONCLUSIONS

Paredrine and epinephrine were given intramuscularly to test the validity of the ballistocardiographic method of determining output. Computed output following administration of these drugs gave results identical with those obtained by generally accepted technics.

Isuprel was given sublingually, by aerosol inhalation or by subcutaneous injection to

thirty-six normal subjects. Consistent increases in cardiac output and pulse pressure were noted after subcutaneous administration. Less marked effects occurred after sublingual administration and none followed aerosol inhalation.

Cardiovascular effects following aerosol administration were minimal. Cardiovascular changes following the administration of isuprel subcutaneously may be as marked as those produced by epinephrine.

Acknowledgment: Isuprel was supplied by courtesy of Winthrop-Stearns, Inc. and paredrine was supplied by courtesy of Smith, Kline & French Labs.

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Re-evaluation of the Reliability of Pharmacologic and Cold Pressor Studies in Hypertension and Pheochromocytoma*

JAMES A. EVANS, M.D., HYMAN J. RUBITSKY, M.D., CARL C. BARTELS, M.D. *and* ELMER C. BARTELS, M.D.

Boston, Massachusetts

With physicians becoming more aware of the importance of including pheochromocytoma in the differential diagnosis of hypertension and of paroxysmal episodes associated with either fixed or labile hypertension,[1–5] the choice and reliability of diagnostic agents for this frequently curable condition become increasingly significant. Since the usual hypertensive syndromes comprise the vast majority of cases and since pheochromocytoma is relatively rare, judgment based on false positive or false negative results of pharmacologic tests is likely to be regrettable. We are constantly faced with this consideration on the hypertensive service at this clinic and consequently have reviewed some of our experiences over the past four years. This report will be concerned only with the use and interpretation of diagnostic drugs and cold pressor studies since numerous papers dealing with the clinical syndromes are constantly appearing in the literature.

There are two types of agents to be discussed. The first type is that which induces overshoots of hypertension, with or without an ensuing clinical paroxysm, which may be used in patients whose blood pressure is normal or elevated. The second type is the "adrenergic-blocking" agents used in patients with sustained hypertension which significantly lower blood pressure in a way considered diagnostic of pheochromocytoma. In the first category we have used histamine, mecholyl® and etamon;® the adrenergic-blocking agents employed were dibenamine® and benzodioxane. Particular attention was directed to determining which of these agents could be used most reliably as a "screening" test.

The technic of administration of these agents is that generally advocated. The patient rested until basal blood pressure readings were obtained. In two patients with pheochromocytoma in whom it was impossible to obtain constant basal readings the studies were carried out under intravenous pentothal® or rectal avertin anesthesia. In the great majority of histamine, mecholyl® and etamon® tests blood pressure readings were taken at one-half to one-minute intervals and were followed for at least thirty minutes. Since we have encountered several delayed true positive reactions, one at thirty-two minutes, we believe that blood pressure readings should be taken for at least one-half hour following a provocative test. Blood pressure readings were taken at two-minute intervals for sixty minutes during the administration of dibenamine and at ten-minute intervals thereafter for two to four hours. The usual doses employed were as follows: (1) histamine phosphate, 0.025 to 0.05 mg. of the base intravenously within several seconds;[6] (2) mecholyl (methacholine chloride), 10 mg. subcutaneously (which dose we have adopted instead of the 25 mg. dose originally advocated[7] because of the lesser incidence of severe side reactions while still maintaining the relative specificity of the test); several studies were performed with doses of 15 to 25 mg.; (3) etamon (tetraethylammonium chloride), 300 mg. intravenously within several seconds; (4) dibenamine, 5 mg. per kg. of body weight administered as a saline infusion over a period not longer than sixty minutes; total doses ranged from 182 to 440 mg., and (5) piperidylmethyl-benzodioxane (933 F), 0.25 mg. per kg. of body weight given over a

* From The Lahey Clinic, Boston, Mass.

two-minute period through a side tube following stabilization of blood pressure during an infusion of saline solution. We have not employed this agent as extensively as dibenamine® because of the frequent hypertensive and other side reactions encountered with its use in patients not having pheochromocytomas. Also, since pheochromocytomas are so rarely seen clinically, we wished to concentrate most of our experience on one adrenergic agent. Moreover, excellent reports dealing with benzodioxane are already in the literature by Goldenberg and Aranow.[8]

TABLE I

CLINICAL DIAGNOSES IN CASES OF PSEUDOPHEOCHROMOCYTOMA (MADE IN ADDITION TO HYPERTENSION)

Group II	
Anxiety state or hysteria	10
Convulsions (etiology undetermined)	2
Malignant hypertension	2
Diencephalic syndrome	2
Cystic lesion of the kidney	1
Periadrenal, non-specific inflammatory mass	1
Menopausal syndrome	1
Persistent hypertension after sympathectomy	1
Chronic pyelonephritis	1
Epilepsy	1
Migraine	1
Brain tumor.	1

At all times during the studies epinephrine, adrenolytic agents, atropine and pitressin were available as antidotes.

The case reports fall into three groups as follows:

Group I consists of 250 consecutive patients who were subjected to bilateral lumbodorsal sympathectomy (usually the fourth thoracic to the second lumbar segment) and splanchnicectomy for essential or malignant hypertension during the past four years. None of these patients was found to have adrenal cortical tumors during routine exploration of the perirenal area by our neurosurgeons.

Group II comprises twenty individuals with either labile or relatively fixed hypertension in whom the diagnosis of pheochromocytoma was entertained strongly enough at some point to merit pharmacologic tests or exploratory surgery. (Table IX.) Eight exploratory operations were performed in this group without finding a pheochromocytoma although it is conceivable that aberrantly-located chromaffin tissue might have been missed. In seven of these a first stage splanchnicectomy was performed at the same time and later followed with a second stage. It was thought from the studies carried out that no functioning pheochromocytoma was present in any of the remaining twelve patients. The final diagnoses in these cases are summarized in Table I.

TABLE II

	No. of Cases	Males	Females	Age (yr.)	Duration of Hypertension	Operations
Group I	250	122	128	10–55	1 mo. to 24 yr.	250*
Group II	20	10	11	20–54	5 mo. to 20 yr.	8†
Group III	4	0	4	20–30	5 mo. to 21 yr.	4‡

*Bilateral lumbodorsal sympathectomies.
†Eight explorations followed in 7 cases by first stage sympathectomies.
‡Adrenalectomies.

We encountered no instances of the following conditions which at times are said to be associated with paroxysmal hypertension:[9,10] glomerulonephritis, lead poisoning, tabes dorsalis, vascular brain damage, the hypothalamic syndrome and neuroblastoma. On several occasions the diagnoses of hyperinsulinism, psychomotor epilepsy, postural hypotension, carotid sinus hypersensitivity, hyperthyroidism, Cushing's syndrome, peptic ulcer and periarteritis nodosa were considered but not subsequently confirmed. Page's type of diencephalic hypertension is especially likely to suggest the diagnosis of pheochromocytoma.[11]

Group III consists of our four clinically, surgically and pathologically proven cases of pheochromocytomas, recently reported in detail by Bartels and Cattell.[1] (Table X.)

Table II shows the sex, age range and known duration of the hypertension. It will be seen that whereas males almost equaled females in groups I and II all our patients with pheochromocytoma in group III were young females. It is of interest that the incidence of males in group I was not greater since there is an apparently greater incidence of malignant and progressive hypertension in middle-aged males. It is evident that sex incidence in a small series does not help in the differential diagnosis of pheochromocytoma and other types of paroxysmal hypertension because in larger series of patients with pheochromocytoma the sex incidence is equally

divided.[2] The almost equal division of sex in group II offers no aid in differential diagnosis although the predominance of cases of anxiety and hysteria associated with paroxysmal hypertension might ordinarily lead one to expect a larger proportion of females than was found.

TABLE III

	No. of Cases	Retinopathy					Cardiomegaly or Angina	Impaired Renal Function	Cerebral Accidents
		0	1	2	3	4			
Group I	250	0	36	100	84	30	98	48	41
Group II	20	1	3	13	2	1	5	4	2
Group III	4	3	0	0	1	0	2	0	1

In Table III the distribution and frequency of the significant aspects of the patient's clinical hypertensive state are summarized as regards pathologic changes in the retina, the cardiac-coronary status, kidney function (non-protein nitrogen, phenolsulfonphthalein, urea clearance and intravenous pyelograms) and cerebral circulation.

COLD PRESSOR STUDIES

The results of cold pressor studies in these three groups are summarized in Table IV. They were carried out either at the clinic or in hospitalized patients as part of our work-up to determine the advisability of sympathectomy. The test was performed only after a rest period in the reclining position for at least fifteen minutes and in patients not given sedatives and to whom the details of the test were fully explained. The hand was immersed in ice water for one minute and blood pressure readings were taken at one-half to one-minute intervals from the onset. In addition to taking the usually accepted rise of 20/15 mm. as indicative of "hyperreactors" we have further presented the "hyporeactor" rises as being either less than 10/10 or less than 20/15 but greater than 10/10 mm. A rise of either systolic or diastolic pressure above 20/15 placed the patient in the higher group.

TABLE IV

COLD PRESSOR STUDIES

	No. of Tests	$<\frac{10}{10}$	$<\frac{20}{15}$	$>\frac{20}{15}$	Hyperreactors (Per cent)
Group I	184	37	51	96	52
Group II	17	6	2	9	53
Group III	3	0	0	3	100

The reasons for presenting these data are twofold. First, we have arbitrarily taken the degree of blood pressure rise on the cold pressor test as the level to be exceeded by the "overshoot" in the various provocative tests for pheochromocytoma before we consider the blood pressure rise *per se* to be significant. Roth and Kvale[12] and Aranow[9] have also adopted this policy. Second, our experience differs from that of Smithwick who believes that a normal or hyporeactive response to cold is decidedly the exception in essential hypertension (ten of his one hundred cases) whereas it is commonly observed in patients with pheochromocytomas (seven of his nine cases[2]). We have found a 50 per cent incidence of "hyporeactors" (that is, less than 20/15) in group I but no "hyporeactors" in our patients with pheochromocytomas. In fact, in the three studies in group III, including one performed under anesthesia, the patients were marked "hyperreactors."

MECHOLYL STUDIES

We have had extensive experience with mecholyl® over the past four years as a screening test for all types of hypertensive patients and particularly for those in whom the diagnosis of pheochromocytoma is entertained. In their initial report of the use of this test Guarneri and Evans[7] repeatedly produced a considerable "overshoot" (plus 106/60) with 25 mg. of mecholyl® in a patient with a pheochromocytoma. This occurred whether the initial hypotensive reaction occurred or was blocked by atropine, or whether the nicotinic action of mecholyl® at the preganglionic sympathetic-

adrenal synapse was blocked by curare given in a dose large enough to produce muscular and respiratory paralysis. These observations, in addition to those to be presented herein dealing with the effects of anesthesia in blocking the response, lead us to the conclusion that the specificity of the test is related to a direct cellular stimulation of the chromaffin tissue in the tumor with subsequent release of either epinephrine or nor-epinephrine.

TABLE V

MECHOLYL STUDIES

	No. of Tests	Overshoot		False Negative
		< Cold Pressor	> Cold Pressor	
Group I	110	1*	0	--
Group II	19	1†	1‡	--
Group III	5	1§	4	0

*Overshoot plus 10/5 (up to 230/145); 4-hr. blood pressure, high 260/140; cold pressor, plus 30/10.

†Overshoot plus 20/20 (195/135); 4-hr. blood pressure, high 246/150; cold pressor, plus 45/20.

‡Overshoot recorded only at 90 min. plus 20/10 (170/110); 4-hr. blood pressure, high 200/120.

§Overshoot plus 10/20 while under anesthesia; overshoot of plus 30/20 without anesthesia; cold pressor, plus 30/15.

The results of our mecholyl studies are presented in Table v. If one uses the term "overshoot" to mean any blood pressure reading exceeding the basal resting controls at the onset of a test, it can be seen that the test has over 98 per cent reliability in the diagnosis of pheochromocytoma. Furthermore, if one accepts as significant only overshoots greater than the cold pressor rise which occurs within thirty minutes after the onset of the mecholyl® test, and in non-anesthetized patients, the test assumes an accuracy of almost 100 per cent. This statement appears to be warranted from the following facts which may also be gleaned from Table v: (1) Two of the three "false positive" tests (groups I and II) were less than the cold pressor rise. (2) The third "false positive" was recorded only at ninety minutes after mecholyl® had been given. No overshoot had occurred within the first hour of the study. (3) No false negative tests occurred in the eight patients in group II whose adrenal glands were examined at the time of surgery. (4) The only positive test among the pheochromocytoma patients which was less than the cold pressor rise was obtained under anesthesia. This individual when not under anesthesia exhibited an overshoot greater than the cold pressor test.

It should be noted that we have not presented the many negative mecholyl tests done in the now routine work-up of hypertensive patients who have not been candidates for surgery. Early in our experience with the mecholyl test we gained such faith in its specificity that six patients with positive tests to histamine or etamon® or both but negative tests to mecholyl were spared exploration. (Table IX.) To be sure, none of these patients had other evidence of pheochromocytoma that would have compelled us to advise exploration.

The test has not been done in patients with bronchial asthma or angina pectoris. The fact that no serious side effect has occurred in over four years (the test having been performed by more than ninety different physicians during that time) leaves little further to be said concerning its safety when the aforementioned precautions are taken. With the 10 mg. instead of 25 mg. dose now routinely employed the sweating, salivation, tearing, dyspnea, flushing, warmth and urge to defecate or urinate are transient and tolerable, rarely requiring termination of the test with atropine.

With regard to the specificity of the test, it is of interest that Cases 1, 2 and 3 in group III reverted to negative responses postoperatively whereas Case 3 still evidenced some histamine hyperreactivity postoperatively. In Case 1 after the patient had remained well for one year following removal of her pheochromocytoma, a swelling developed in the right side of the neck which proved by biopsy to be a metastatic malignant pheochromocytoma histologically similar to the one removed at operation. The blood pressure had remained normal with no further hypertensive attacks for three and a half years; the mecholyl® test was negative. We have not encountered a functioning metastatic pheochromocytoma after removal of the primary tumor, as has recently been described.[13]

We have reviewed our material very closely to detect any false positive tests to mecholyl and have been unable to do so. It is conceivable that difficulty might be encountered in evaluating an "overshoot" in patients with very labile hypertension and in whom the control reading is taken when a very low blood pressure

is present. If on mecholyl test subsequent pressures are recorded above this level, but not exceeding the average blood pressure in that patient, the question of whether a real overshoot actually occurred might arise. This problem presented itself in only one patient with a strong family history of hypertensive disease who probably did not have a pheochromocytoma but anxiety spells with a labile blood pressure. Shapiro et al.[14] have recently reported a case of pheochromocytoma in which no overshoot to 20 mg. of mecholyl® was observed. However, the end point was recorded at twenty minutes after the administration of the drug, at which time the systolic pressure had reached the control blood pressure level.

TABLE VI

HISTAMINE STUDIES

	No. of Tests	Overshoot		False Negative
		< Cold Pressor	> Cold Pressor	
Group I	7	0	2	--
Group II	12	3	6	--
Group III	5	0	2	3*

*Study in Case 3 done under anesthesia (intravenous pentothal). An overshoot >cold pressor obtained when repeated without anesthesia. Two studies made in one patient (Case 2) under rectal avertin anesthesia. No response to 0.06 mg. of histamine, even though significant hypotension was produced. Etamon and mecholyl tests under anesthesia were both positive.

HISTAMINE STUDIES

Our results with the histamine provocative test are presented in Table VI. It can be seen that there were two significant overshoots in seven studies performed in group I (29 per cent). More striking, however, was the occurrence of nine overshoots (75 per cent) in our diagnostic problem cases in group II, of which there were rises greater than the cold pressor response in six cases (50 per cent) which by our criterion would be called positive tests.

Of a total of five histamine studies in group III there were three false negative responses. Two were performed on the same patient (Case 2) under rectal avertin anesthesia because of the extreme variability of the blood pressure and inability to obtain basal readings of sufficient duration to permit performance of specific diagnostic tests. The dose in both instances was purposely increased to 0.06 mg. and in both instances a good hypotensive effect was obtained. The one etamon® test and two mecholyl® tests under identical circumstances were both significantly positive.

The third false negative test to histamine in group III (Case 3) was also obtained with the patient under intravenous pentothal® anesthesia. The test was positive, however, when no anesthesia was employed. This leads us again to think that the action of histamine, like that of mecholyl,® is probably a direct cellular effect upon the chromaffin tissue which anesthesia and cellular blocking agents may abolish but which synaptic and myoneural junction blocking drugs cannot. It is apparently not dependent to any great extent upon a "sympathetic rebound" resulting from the initial hypotensive response since the initial fall still occurs with atropine or under anesthesia. Sublingual nitroglycerine (0.3 to 1:2 mg.) has been employed as a provocative test without much reliability or specificity;[13] in fact, amyl nitrate has successfully relieved the distressing symtoms of a positive histamine test.[15] These observations are contrary to those of Roth and Kvale who stated that sedation does not have an inhibitory effect on the histamine test.[12] We have not observed a hypertensive episode resulting *per se* from the administration of sodium pentothal,® as has been described.[13] Feldberg[16] and Szczygielski[17] have both shown that histamine is capable of directly releasing epinephrine from the adrenal medulla. Rubitsky et al. have discussed the importance of the "histamine-sympathin see-saw" in allergy and bronchial asthma.[18] We are not aware of any reports concerning the effect of "antihistamine" agents as they modify a positive histamine response in pheochromocytoma.

ETAMON STUDIES

The use of tetraethylammonium bromide was originally advocated as a diagnostic test for pheochromocytoma by LaDue, Murison and Pack.[19] They employed 100 mg. intravenously. It was thought that the use of this agent had an advantage over the other provocative tests in that dangerously high blood pressure levels could be controlled simply by having the patient sit or stand.

Our results with the use of 300 mg. intravenously are summarized in Table VII. It can be seen that of 218 studies performed in groups I

and II there were sixteen overshoots (7.3 per cent), of which one-half were positive by our criterion (that is, overshoots exceeding the cold pressor response). Three patients in group I (the highest clinically recorded blood pressure being 255/130) had levels exceeding 300 mm. for at least four minutes after the injection of the etamon.® Four of these patients also hyperreacted to histamine. All had negative mecholyl® tests.

TABLE VII

ETAMON STUDIES

	No. of Tests	Overshoot		
		< Cold Pressor	> Cold Pressor	False Negative
Group I	204	5*	5	--
Group II	14	2	4	--
Group III	2	0	1†	1‡

*Four overshoots were only in the diastolic pressure with a concomitant systolic hypotension.

†During rectal avertin anesthesia (Case 2).

‡During rectal avertin anesthesia (Case 3).

The two etamon studies in group III were performed under rectal avertin anesthesia. One was negative (Case 3). In Case 2 an immediate overshoot of plus 70/70 was spontaneously followed with a moderate drop and then an overshoot of plus 90/90 which was reduced by having the patient sit up; the blood pressure continued to fluctuate to high levels over the ensuing thirty minutes. Console, Dunbar and Ray[13] have recently called attention to this alternating hypertension and hypotension with associated tachycardia and marked ventricular arrhythmia following the administration of 200 mg. of tetraethylammonium to a patient with pheochromocytoma. This response is in contrast to the usual monophasic hypertensive response and bradycardia to other positive provocative tests. The work of Hoobler, Moe and Lyons[20] suggests that this response is due to blocking of the compensatory responses by the autonomic nervous system which normally "buffers" a rise in blood pressure.

These experiences have been independently borne out by Roth and Kvale.[12] They stated, "We have used this drug on 4 patients with proved tumors. In 2 there was little or no rise in the blood pressure in spite of the existence of a tumor; in 2 others there was a definite rise in the blood pressure. In 2 other cases a rise in blood pressure followed injection of tetraethyalmmonium bromide but the other tests gave negative results and no tumor was found at operation."

TABLE VIII

DIBENAMINE STUDIES

	No. of Tests	Blood Pressure Drop >10/10
Group I	5	1
Group II	14	6*
Group III	1	1

*Several patients became drowsy or fell asleep during the study, at which time the drop was recorded.

We believe that a good hypotensive effect from etamon® is a fair preoperative lability guide to the results that may be obtained by sympathectomy for hypertension and should be used with that purpose in mind. Any overshoots should only raise the suspicion of pheochromocytoma and other pharmacologic tests should be applied. Caution should be taken in any patient suspected of having a pheochromocytoma who is given the drug.

DIBENAMINE STUDIES

We have administered dibenamine® as a saline infusion containing 5.0 mg. per kg. of body weight rather than the 7.0 mg. per kg. advocated by Spear and Griswold.[21] Our data are summarized in Table VIII. We have arbitrarily considered as being suggestive any drop in blood pressure exceeding the basal readings by 10 points in either the systolic or diastolic readings during the infusion. The only significant side effect we have observed has been stimulation of the central nervous system in two patients who received 440 and 330 mg. The postural hypotension, nausea and blurring of vision were not very troublesome. Anuria has not been observed in our experience.[22]

TABLE IX

GROUP II — CASE SUMMARIES

Case, Sex & Age*	Duration of Hypertension (yr.)	B/P Range	Amytal (Low)	Clinical Data	Retina	Heart†	Kidney†	Cold Pressor	Histamine Drop	Histamine Over	Mecholyl Drop	Mecholyl Over	Etamon Drop	Etamon Over	Dibenamine Drop (Lying)	Comment
1, F (54)	20	160–230 110–150	100/60	Recurrent convulsions with hypertension; craniotomy, sugar studies, arteriograms normal	1	E	D	+60 +20	-20 +10	+50 +40	-68 -38	0	-35 -25	+60 +20	-36 -28 (280 mg.)	Negative exploration; sympathectomy
2, F (45)	1 1/2	186–192 108–116	—	Recurrent paroxysms of sweating, palpitations, headaches and nervousness	2	N	N	+10 + 8	0	+52 +28	-52 -38	0	—	—	—	No operation; anxiety state
3, M (36)	1 1/2	124–220 90–118	134/100	Recurrent attacks of headaches, nervousness, profuse sweating, nocturia and dyspnea	3	N	N	+ 6 +24	-24 -30	+40 +20	-24 -34	0	- 40 -110	0	—	Negative exploration; sympathectomy
4, F (27)	3	170–190 110–120	110/70	Recurrent fainting spells every 1 to 2 wk. with anxiety and sweating	2	N	N	+20 +10	-20 -10	+22 + 8	-30 -30	0	-40 -10	0	—	Negative exploration; sympathectomy
5, F (19)	1/2	164–200 110–140	140/100	Headaches at intervals all her life	2	N	N	+8 +6	—	—	-42 -20	0	-26 - 6	-32 + 4	-0 -0 (220 mg.)	Negative exploration; sympathectomy
6, M (48)	4	140–190 60–120	135/70	Recurrent headaches and hypertension precipitated by emotional factors or occurring spontaneously	3	N	N	+20 +12	-25 -30	+40 +20	-40 -30	0	-16 -18	-20 + 2	—	Sympathectomy; negative exploration
7, M (44)	4	235–270 140–148	—	Spells of anxiety and palpitations; "cystic lesion" about left kidney by intravenous pyelograms	2	N	N	+25 +10	—	—	-30 -16	0	-30 - 5	-35 +13	-0 -0 (420 mg.)	Anxiety state; normal retrograde pyelogram
8, M (44)	5	160–210 110–115	150/100	Suspected brain tumor and secondary cerebrovascular accident	4	N	N	—	—	—	—	—	—	—	-40 (dozing) -15 (380 mg.)	Malignant hypertension
9, F (34)	15	140–180 80–110	—	Spells of weakness, chest pain, pallor, flushing ± exciting factors	1	N	N	—	—	—	-25 -15	0	—	—	-0 -0 (260 mg.)	Anxiety state
10, M (45)	4	120–200 110–120	120/80	Spells of jitteriness, trembling and palpitation; diabetic-type glucose tolerance test	0	N	N	(1) - 4 +12 (2) -10 -10	—	+20 +18	-30 -30	0 at 6 min. +20 +10 at 90 min.	-80 -50	0	-76 -32 at 70 min. (388 mg.)	Anxiety state; operation refused
11, F (44)	2	140–186 96–100	—	Attacks of smothering, fainting, chest pain, palpitation and hypertension ("212")	1	N	N	—	-20	0	-35 -14	0	—	—	-20 - 0 at 28 min. -36 - 6 at 45 min. -40 -10 at 75 min. (260 mg.)	Hysteria

12, M (38)	1 1/2	120–215 70–140	—	Palpable left flank tumor; tumor outside upper pole of left kidney	2	N	D	—	−15 +5	−5 +10	−50 −35	0	—	—	−0 −0 (250 mg.)	Periadrenal non-specific inflammatory mass
13, M (46)	9	210–135 (lying) 108–85 (standing)	—	Anxiety and persistent hypertension after a previous sympathectomy	2	N	N	+30 +46	—	—	−50 −30	0	−35 −15	0	−20 −10 (400 mg.)	Refused further surgery
14, F (39)	5	164–186 100–112	—	Recurrent anxiety, weakness, headaches, tremor, dizziness and flushing	2	N	N	+4 +4	(1) −52 −28 (2) −20 −8	0 0	−50 −38	0	−164 −100	0	−0 −0 (200 mg.)	Anxiety state and menopause
15, F (20)	1 3/4	165–200 105–125	—	Hypertension with petit mal type of seizures; normal electroencephalogram	1	N	N	+35 +25	—	—	−20 −30	0	−60 −20	0	−0 −0 (300 mg.)	Probable petit mal
16, M (38)	8	158–182 106–122	—	Spells of numbness, bloating, weakness, headaches, choking and hypertension (220) past 5 mo.	2	N	N	+26 +18	−14 −10	+2 +6	−28 −18	0	−10 −8	−10 +16	−0 −0 (330 mg.)	Anxiety state; migraine
17, M (45)	1/2	180–240 110–150	180/100	Personality changes, mental confusion, recent hypertension, papilledema without hemorrhage or exudate; normal ventriculograms and brain studies	4	E	N	—	—	—	−35 −24	0	—	—	−8 −10 at 30 min. −25 −15 at 60 min. (dozing) (400 mg.)	Malignant hypertension, stabilized with diet and veriloid
18, F (47)	3	140–204 100–140	130/90	Spells of tension, nervousness, giddiness, weakness, nausea, "blacking out"	2	N	D	(1) +8 +10 (2) +2 +4	—	—	−50 −26	0	(1) 0 +14 +16 (lying) +2 +10 (sitting) (2) 0 +12 +18 (lying) −10 +36 (sitting)		—	Left chronic pyelonephritis; 1 cc. of epinephrine caused rise (+48/+26); negative exploration; sympathectomy
19, F (34)	5	160–210 80–140	180/100	Spells of palpitation, shakiness, weakness, sweating, panic, blushing; basal metabolic rate +24, +26, +44	1	E	N	+40 +20	—	—	−54 −36	0	−60 −110	0	—	Anxiety state
20, F (39)	5	135–246 98–150	130/95	Spells of sweating, hot flushes, pressure, headache, palpitations; insensitive to 1 cc. of epinephrine; normal glucose tolerance test and basal metabolic rate	2	E	N	+45 +20	−25 −16	+25 +12	−35 −27	+20 +20 (High (195 135)	—	—	−0 −8 (220 mg.)	Careful exploratory negative; sympathectomy at later date

*Figures in parentheses represent age.

†N = normal; E = enlarged; D = diminished.

It can be seen that there were seven false positive tests in nineteen studies made in groups I and II (37 per cent). These patients for the most part had sustained hypertension and we were not inclined to give them benzodioxane because of the alarming hypertensive reactions which we and others have encountered with its use.[22,23] Most of these drops in blood pressure with dibenamine® were not profound. We believe that several of these hypotensive responses probably were related more to the patients being reassured and relaxing (two actually falling asleep) during the prolonged infusion than to any specific pharmacologic action of the drug. Aranow[9] and Wunsch, Warnke and Myers[24] have found that a depressor effect may be observed with this drug in normal individuals and in patients with hypertension resulting from causes other than pheochromocytoma.

On the other hand, we were helped considerably several times by a negative dibenamine® test in cases of histamine and etamon® hyperreactors and particularly in the one patient with a false positive overshoot to both histamine and mecholyl® after ninety minutes (Cases 7, 10, 12, 16 and 20). (Table IX.)

Dibenamine was used as a diagnostic aid in Case 4 of the pheochromocytoma series. After basal blood pressure readings of 190/120 had been obtained, the infusion was begun. Within fifteen minutes the systolic pressure was 140; and when the test was completed at the end of an hour (after 182 mg. had been given), the blood pressure was 120/80. Based on our experience with dibenamine® in only one actual case of pheochromocytoma we would regard this test as positive only if the blood pressure is reduced to normal or very near normal levels. We have not yet had the opportunity of preventing the pressor effect of the provocative tests with dibenamine,® as reported by Spear and Griswold.[21] This blocking effect of dibenamine® would be the ideal diagnostic demonstration of pheochromocytoma and, of course, is the only diagnostic way of using dibenamine® in patients with pheochromocytomas of the paroxysmal type.

Pitcairn and Youmans[25] have clearly shown that the dibenamine® test can be greatly influenced by the relative preponderance of epinephrine and nor-epinephrine present in removed pheochromocytomas. Dibenamine® failed to block the pressor activity of nor-epinephrine in dosages which were considerably greater than those necessary to block and reverse the pressor response to various doses of epinephrine. Several other adrenergic-blocking agents (933 F, yohimbine, ergotamine and ergotoxine) are also known to be less effective in blocking the pressor activity of nor-epinephrine than of epinephrine.[26,27] One cannot determine by the response of the arterial blood pressure to dibenamine® either the nature or the amounts of pressor substance (s) which are liberated into the circulation from pheochromocytomas.[25]

Dibenamine® was successfully used during the surgical removal of the tumor from Cases 3 and 4.[1] The blood pressure was purposely not completely restored to normal—only doses of 60 and 175 mg. being used, respectively—for fear that profound hypotension might ensue if too much were given. This fear has been dispelled by the gratifying experience of other investigators with priscoline,[13] piperoxan,[15] dibenamine[13] and C-7337[28] prior to and during removal of these tumors, as a result of which both the initial hypertension and the subsequent hypotension have been prevented to a considerable degree. This would appear to suggest a cardiac basis for the hypotension and to disprove the concept that the hypotension results from removal of the tumor *per se* and that replacement therapy with epinephrine and other vasoconstrictors is indicated. One of us (E. C. B.) has suggested that the fall in blood pressure and cardiac failure met so often on removal of a pheochromocytoma or in patients with pheochromocytomas submitted to other surgical procedures are the result of an epinephrine-produced "clamp" of the large vessels proximal to the brachial artery, and the emergency use of epinephrine is contraindicated.

There was no operative mortality in our pheochromocytoma series.

PIPERIDYLMETHYL BENZODIOXANE (933 F)

We obtained no false positive tests with this agent in three puzzling pseudopheochromocytoma syndromes. Marked pressor responses with symptoms occurred in two patients. This has previously been reported.[3,12,22,23] Symptomatic rebound hypertension of a high order has also occurred following the diagnostic hypotension produced by benzodioxane in patients with pheochromocytoma.[15] The test does have the advantage over dibenamine® in that the infusion itself takes only two minutes so that blood pressure responses are more clear-

TABLE X

GROUP III -- PROVEN PHEOCHROMOCYTOMAS (FROM BARTELS AND CATTELL)

Case, Sex & Age*	Duration of Hypertension	B/P Range	Amytal (Low)	Clinical Data	Retina	Heart†	Kidney†	Cold Pressor	Histamine Drop	Histamine Over	Mecholyl Drop	Mecholyl Over	Etamon Drop	Etamon Over	Dibenamine Drop (Lying)	Comment
21, F (23)	3 yr.	102–94 230–140 (during attacks)	190/132 P, 120	Recurring daily attacks of weakness, headaches, palpitation, dyspnea, nausea and vomiting; thyroidectomy 2 yr. before; basal metabolic rate +36 and blood sugar 166 during an attack	0	N	Left depressed	(B) +32 / +38 (A) +20 / +15	(B) (0.5 mg.) -23 / -35 (A) -40 / -44	+105 / + 68 +40 / +40	(B) -20 / -20 (A) -20 / -15	+100 / + 60 0	---		---	No further attacks after surgery; developed metastatic pheochromocytoma in neck 1 yr. later; mecholyl test negative
22, F (24)	5 mo.	120–80 200–150 (very labile)	120/90‡ P, 110	Persistent hypertension, poor vision, perspiration, flushing and pounding of head since a previous toxemia of pregnancy	3	Pulsus alternans	D	+75§ / +90	(B) (0.06 mg.) -25§ / -25 -15§ / -25 (A) -20 / -15	0 +10 / +25 0	(B) -20§ / -25 0§ -25 / -20	+110 / + 80 +133 / + 90 0	(B) 0§	+90 / +90 (lying) -40 / -30 (sitting)	---	Cure of attacks and hypertension; slight postoperative aphasia
23, F (30)	21 yr.	110–180 80–100	120/80‡ P, 82	Attacks of substernal distress, palpitation, perspiration, heat intolerance and dyspnea; toxemia of pregnancy; diabetic glucose tolerance test; basal metabolic rate +53 and +60 under pentothal anesthesia	0–1	Angina	Rt. suprarenal calcification	(B) +30 / +15 (A) +10 / 0	(B) -50 / -30 ‡ -40 / -40 (A) -10 / -10	+20 / +20 0 +20 / +10	(B) -40 / -30 -70 / -50 (A) -10 / -10	+10 / +20 (at 30 min.) +30 / +20 (at 32 min.) +5 / 0	(B) -50§ / -40	0 ---	60 mg. during surg. dropped B/P from 180/126 to 120/90	Cure of attacks, hypertension and abnormal glucose tolerance test
24, F (20)	8 mo.	190–200 120–130	140/90 P, 120	Continual sweating and palpitation at night; basal metabolic rate +37, +42, +40; diabetic glucose tolerance test	0	N	Left depressed								182 mg., basal 190/120, 15 min. 140/90, 60 min. 120/80	Cure of symptoms, hypertension, abnormal glucose tolerance test and hypermetabolism

*Figures in parentheses represent age.

†N = normal; D = diminished; (B) = before surgery; (A) = after surgery.

‡Intravenous pentothal.

§Rectal evertin.

cut. It is, however, apparently specific only for adrenal medullary tumors associated with *persistent* hypertension. Roth and Kvale have reported on two patients with paroxysmal hypertension and verified tumors in whom only a pressor response was elicited.[12] False positive studies have also been encountered.[28]

The mechanism of action of 933 F is still not clarified. Rosenblueth and Cannon[29] have suggested that it "polarizes" the effector cells, decreases cellular permeability and subsequently limits the penetration of circulating chemicals without affecting the activity of mediators liberated intracellularly. Morison and Lissak[30] thought that it does not act "by rendering the effector cells impermeable" but rather as an accelerator in the inactivation of epinephrine. Finally, Calkins et al.[10] believed that benzodioxane depends for its effectiveness in blocking the pressor effects of epinephrine on a competition between it and the pressor amines for a specific cellular epinephrine receptor.

COMMENTS AND SUMMARY

An attempt at re-evaluation of the reliability of pharmacologic and cold pressor studies in hypertension has been made. This is based on the response of 250 consecutive patients subjected to sympathectomy for hypertension (group I), twenty patients with either labile or fixed hypertension in whom the diagnosis of pheochromocytoma was considered at some time (group II) and four clinically and pathologically verified cases of pheochromocytoma (group III). (Table X.)

Many patients in group II were females with anxiety states or hysteria and labile hypertension. All our pheochromocytoma cases were young females. Since patients in the group of pseudopheochromocytoma syndromes may present an almost complete "textbook picture" of pheochromocytoma clinically, we believe that no patient should be subjected to exploratory operation without first having an unequivocally positive response to one of the pharmacologic tests, especially mecholyl.

The *cold pressor response* is of importance because we believe that the blood pressure "overshoot" to any of the provocative tests must exceed it before the blood pressure rise from the provocative tests can be considered significant. Contrary to other reports, we found an incidence of only 50 per cent "hyperreactors" in essential hypertensive patients (group I), and of 100 per cent in our patients with pheochromocytoma.

Mecholyl® (*methacholine chloride*) as a 10 mg. dose subcutaneously is, in our experience, the best, safest and simplest screening provocative test for all hypertensive or normotensive patients in whom the diagnosis of pheochromocytoma is entertained. In our experience the test has been both reliable and specific if one accepts as significant only those "overshoots" greater than the cold pressor rise which occur within thirty minutes after the onset of the test and in a non-anesthetized patient. In group I there were 29 per cent false positive histamine tests. In our cases, which were diagnostic problems (group II), there were 50 per cent false positive histamine tests and three more patients showed overshoots less than the cold pressor test.

Of a total of five histamine studies done in group III there were three negative tests, all of which were performed under anesthesia. Three of the four patients with pheochromocytoma had positive tests to histamine when anesthesia was not used.

In two of the patients with pheochromocytoma anesthesia had to be used to obtain a basal blood pressure. It should be remembered, however, that anesthesia may diminish or abolish an otherwise positive provocative test.

It is thought that mecholyl and histamine produce their positive results in pheochromocytoma by a direct cellular effect upon the chromaffin tissue which anesthesia and cellular receptor blocking agents may abolish but which synaptic and myoneural junction blocking drugs cannot.

There were 7.4 per cent false positive responses to 300 mg. of *etamon* intravenously. Of the two tests made under anesthesia in patients with pheochromocytoma one was positive and one was negative. A characteristic fluctuating hypertension-hypotension response to etamon,® described by Console et al.,[13] was observed in pheochromocytoma (Case 2).

Dibenamine,® administered as a saline infusion containing 5.0 mg. per kg. of body weight, has been of considerable diagnostic help in certain patients with sustained hypertension. There was, however, a 37 per cent incidence of probable false positive tests. Dibenamine® can also be employed to good advantage for the following: (1) as a check on positive provocative tests by observing whether the "overshoot" can be blocked; (2) to evaluate the effect on paroxysms

clinically and (3) to prevent the hypertensive crisis with subsequent acute left heart failure and peripheral hypotension during the operative manipulation and removal of the tumor.

In group II there were two patients with very labile blood pressures and paroxysmal types of blood pressure with negative provocative tests whose paroxysms were abolished for as long as two weeks, with marked symptomatic improvement. This further shows the non-specificity of dibenamine® as a diagnostic test for pheochromocytoma. Dibenamine® has the added disadvantage of a slower onset of action and a more prolonged adrenolytic action than the imidazolines.

Benzodioxane has proved of value in three cases in group II but is limited to some extent by the pressor response occurring not only in the usual hypertensive syndromes but also in paroxysmal hypertension due to pheochromocytomas. Just as the clinical picture of pheochromocytoma may be greatly influenced by the relative and absolute amounts of epinephrine and norepinephrine present, so the results of studies employing adrenolytic agents may vary. Not only is arterenal about one-tenth as active in raising the blood sugar level as epinephrine and considerably less active on the heart than on the peripheral vessels, but also its pressor action is much less affected by adrenolytic drugs than that of epinephrine.[31]

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Review

Pharmacologic Basis of Various Tests Used in the Diagnosis of Pheochromocytoma*

GEORGE ENTWISLE, M.D., CLEMENT A. STONE, M.SC.† *and* EARL R. LOEW, PH.D.
Boston, Massachusetts

WITHIN recent years the number of reports and review articles on pheochromocytoma has progressively increased, especially since the advent of various tests used as diagnostic aids. The clinical picture of paroxysmal hypertension and associated symptoms characteristic of the syndrome is well known although many cases are being detected with persistent hypertension. Prior to 1945 the diagnosis of suspected pheochromocytoma was based largely on the attacks which characterize the clinical picture, attacks occurring either spontaneously or precipitated by various means such as abdominal massage, changes in position or physical exertion. Visualization of the tumor by roentgenologic examination with or without perirenal air insufflation and the presence of a pressor substance in the blood were other demonstrations of diagnostic value.

In 1945 Roth and Kvale introduced the intravenous use of histamine as a diagnostic test for pheochromocytoma.[1] They found that this agent induced typical attacks in patients with this disease. Since that time other pharmacologic agents, such as tetraethylammoniumbromide (TEA®), mecholyl,® benzodioxane® and dibenamine,® have been used as testing substances to assist in the diagnosis of this condition. Since the pharmacologic basis for the action of a few of these substances is either occult or not well known, we were prompted to review the available literature and conduct animal experiments in an attempt to clarify the action of these drugs.‡

In general the agents used as testing substances in cases of suspected pheochromocytoma can be divided into two groups: Those in the first group are substances which cause or produce a typical attack. This type of agent is especially useful in the paroxysmal form of the disease and is best fitted for use between attacks. Second are those substances which block effects of epinephrine and/or nor-epinephrine released spontaneously or following some test procedure. These substances are the adrenergic blocking agents which are especially suited to those cases with persistent hypertension.

Histamine. Histamine was introduced as a test for pheochromocytoma by Roth and Kvale[1] in 1945. Acting on the findings of Hyman and Mencher[2] they found that the intravenous injection of 0.025 to 0.050 mg. of histamine base produced a typical attack in proved cases of pheochromocytoma, with a rise in arterial pressure approximately 100 mm. Hg in excess of the elevation produced by the cold pressor test. In normal patients or former pheochromocytoma patients after removal of the tumor the response was characterized by a rise in pressure somewhat less than that produced by the cold pressor test. Since 1945 the histamine test has been used rather frequently with good success[3–6] although instances of its failure in proved cases have been reported.[5–7]

The mechanism of action of histamine in causing a typical attack is not well known. It was not indicated in the original article[1] and is not discussed in many later articles on pheochromocytoma. Although Hyman and Mencher implied that histamine acted directly on the adrenals,[2] most recent articles explain the response of epinephrine liberation from the tumor as probably reflex in origin due to the fall in blood pressure produced by histamine.[3,8,9] Although there is some experimental evidence for this mechanism,[10] there is fairly convincing

* From the Department of Physiology, Boston University School of Medicine, Boston, Mass.
† Life Insurance Medical Research Student Fellow.
‡ The histamine, mecholyl and tetraethylammonium chloride used in this investigation were kindly supplied by Parke, Davis & Co., Detroit, Mich.

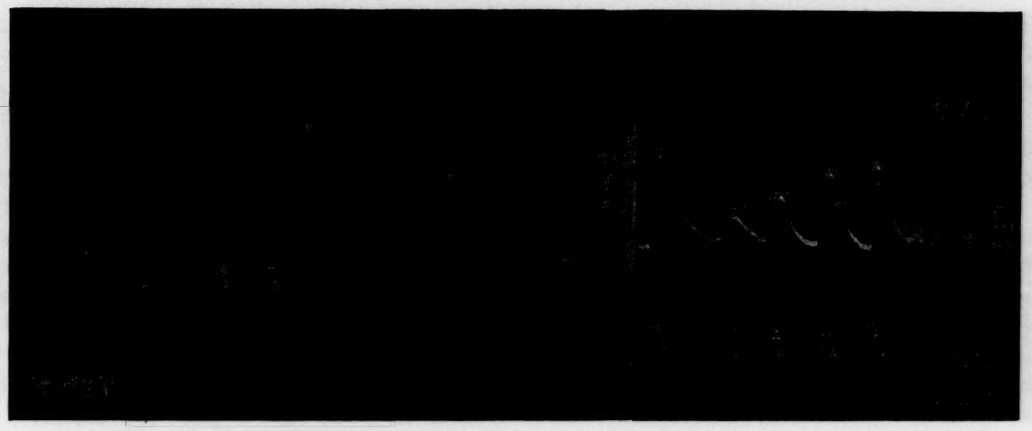
evidence in the literature that histamine can cause epinephrine liberation by direct stimulation of the adrenal medulla. This then would point up two possible mechanisms whereby histamine can act on the adrenal medullary tumor.

One of the earliest convincing investigations showing that histamine causes epinephrine liberation is that of Dale[11] who showed that small doses of histamine produced pronounced dilatation of the sensitized pupil of the cat similar to the dilatation produced by small doses of epinephrine. Kelloway and Cowell[12] also presented evidence that epinephrine was liberated at least in part by direct stimulation of the adrenal medulla.

Subsequently histamine was found to produce epinephrine liberation in isolated perfused glands.[13] In 1926 Burn and Dale,[14] employing decapitated eviscerated cats, showed that small doses of histamine caused marked pressor responses which could be reversed after ergotamine and which were not produced reflexly by equivalent depressor stimulation from other drugs. Their work was subsequently confirmed by Feldberg and Minz[15] in 1931. Using eviscerated cats these workers compared the blood pressure response to intravenous histamine with the response produced by giving the histamine intra-arterially close to the adrenal glands. They found that when small doses were used intravenous histamine caused only a depressor response while intra-arterial histamine caused marked pressor responses, frequently without any prior depressor component. They were unable to cause pressor responses in their preparation reflexly by the drop in blood pressure induced by giving other drugs, usually acetylcholine. They concluded, as have others, that small doses of histamine liberate more than an equivalent amount of epinephrine. Emmelin and Muren[16] confirmed earlier work and demonstrated in cats that small intra-arterial doses of histamine cause contraction of the sensitized nictitating membrane coincident with the sharp rise in arterial pressure. Both of these responses were prevented by prior administration of antihistaminic compounds which prevent this release of epinephrine from the adrenal medulla.

The preparation introduced by Feldberg and Minz[15] is ideally suited to the study of drug action on the adrenal medulla. It consists of an eviscerated animal (usually the cat) in which the inferior vena cava and abdominal aorta are tied below the renal vessels and the renal vessels are tied close to the hilus of the kidney. A small cannula or catheter is placed in the stump of the celiac artery close to the adrenals. For intravenous comparisons another cannula is placed in a peripheral vein. A denervated nictitating membrane can be incorporated into this preparation, or a spinal preparation used, either with or without denervation of the adrenals. The effect of small doses of any drug on the adrenal medulla can be elucidated by the comparison of intra-arterial with intravenous responses. This preparation was used and a total of seventy-two injections of histamine base (0.20 to 2.5 μg.) were given intra-arterially in thirteen cats. There was fairly marked individual variation in the responsiveness to intra-arterial histamine in these cats but in all animals a dose of histamine was found which caused sharp rises of blood pressure when given close to the adrenals. With variation of dosage a total of forty-two pressor responses were produced, averaging 26 mm. Hg and preceded in only thirty-three instances by a depressor phase of an average of 16 mm. Hg. In each of the thirteen cats a comparison was made of the intra-arterial and intravenous responses to the same dose of histamine (0.20 to 0.40 μg.). The average response of the intra-arterial injection was a fall of 10 mm. Hg followed with a rise in pressure of 22 mm. Hg. Somewhat the reverse occurred following intravenous injections since there was a fall in pressure of 32 mm. Hg followed with a rise of 8 mm. Hg. A typical response is found in Figure 1A. Reflex stimulation of the adrenals in these animals was prevented by splanchnicectomy and/or celiac ganglionectomy. The nictitating membrane of three other cats was sensitized by denervation two weeks before the experiment. Histamine in a dose of 0.5 μg. was given intra-arterially on four occasions resulting in a contraction of the denervated nictitating membrane on each occasion. (Fig. 1B.) Intravenous injection of this same dose of histamine caused no contraction of the membrane. These results prove that histamine acts directly on the adrenal medulla to cause the release of one or more pressor substances.

The adrenal medulla is quite sensitive to histamine. The pressor response resulting from the intra-arterial injection of 0.20 to 0.40 μg. of histamine in most experiments was roughly equal in magnitude to the pressor response resulting from 1.0 to 2.0 μg. of epinephrine given



A B

FIG. 1. A, spinal eviscerated cat; artificial respiration; renal vessels ligated at hilus of kidney; inferior vena cava and abdominal aorta tied below renal vessels. Intra-arterial (I.A.) catheter placed in stump of celiac artery for retrograde injections. Intravenous (I. V.) catheter in jugular vein. A = acetylcholine, 5 μg; H = histamine, 0.4 μg; E = epinephrine, 2 μg.; T = tetraethylammonium chloride, 4 mg. B, eviscerated cat; nembutal anesthesia; spontaneous respiration; right nictitating membrane denervated two weeks before experiment. NM = nictitating membrane. Other symbols as in Figure 1A.

intravenously. In Figure 1A can be seen the comparison between the intra-arterial and intravenous injection of histamine. In Figure 1B the sharp pressor response caused by intra-arterial histamine is seen to be coincidental with the contraction of the sensitized nictitating membrane. The intravenous route causes only a depressor response without any contraction of the nictitating membrane. The pressor responses to histamine are due to direct stimulation of the adrenal medulla and not to nervous reflexes since they are obtained in spinal cats after the adrenal glands are denervated by removal of the celiac ganglion.

Tetraethylammonium Bromide (TEA). In 1947 the intravenous use of TEA as a diagnostic test for pheochromocytoma was suggested by LaDue, Murison and Pack.[17] These investigators reported that 400 mg. of this ganglionic blocking drug injected intravenously caused a marked pressor response lasting fifteen minutes. They stressed an advantage of TEA over histamine, namely one of safety, for any pressor response can be controlled by tilting the patient upright. The TEA test has been regarded with favor by some[6,7,18] whereas other investigators[4,5,19] have not had consistent success with it (possibly at least in one case[4] because the dosage used was only 100 mg. of TEA and in another[19] it was not given intravenously). The mechanism of action of this drug is not clear. It has been postulated that the outpouring of epinephrine is due to vasodilation and increased blood flow through the tumor resulting from blockade of the autonomic ganglia.[17] On the other hand, it has been reported that TEA causes either no change or a decrease in the blood flow to the kidneys.[20]

It is known that TEA, especially in high doses, can cause marked pressor responses in experimental animals. This action has been attributed to a direct effect on vascular smooth muscle.[20] Recently Page[21] presented experimental evidence to show that TEA causes liberation of norepinephrine from the liver and to a lesser extent from the adrenals.

The effect of this drug on the adrenals, with use of the preparation of Feldberg and Minz, has been studied experimentally since it is probable that tetraethylammonium ion is capable of directly stimulating the adrenal medulla. TEA in a total dose of 0.5 to 4.0 mg. was given intra-arterially in this preparation on twenty occasions in thirteen cats. In only one case was there no pressor response. The average pressor response was 41.6 mm. Hg. In contrast, the responses to intravenous injection of the same doses of TEA (eighteen injections in thirteen cats) were variable with respect to both occurrence and magnitude. On only five occasions were there pressor responses greater than 10 mm. Hg (19, 16, 18, 22 and 12 mm.). The usual response was a change in blood pressure of less than 8 mm. Hg.

A comparison was made between the intra-arterial and intravenous responses to 4.0 mg.

of TEA in this preparation when incorporated with a denervated nictitating membrane. A total of eleven intra-arterial and eleven intravenous injections were made in seven cats. This dose intra-arterially resulted in a marked pressor response and contraction of the sensitized membrane which averaged 20 mm. in height. The intravenous injection of this dose resulted in little change in arterial pressure and only minimal contraction of the nictitating membrane (average 2.0 mm.) (Fig. 1B.)

The pressor response and membrane contraction must be due to released epinephrine since the responses were blocked by a specific adrenergic blocking drug. Furthermore, they were not obtained after adrenalectomy. In this preparation the liver was not removed; but since the arterial supply to the liver had been completely removed, the possibility that the pressor effects were due to liberation of nor-epinephrine from the liver is extremely unlikely. These animal experiments form a basis for inferring that TEA causes the marked pressor responses or "attacks" in patients with pheochromocytoma by direct stimulation of the adrenal medullary tumor.

Mecholyl. Mecholyl was used by Guarneri and Evans[22] in a case of pheochromocytoma in 1948. They found that the intravenous injection of 25 mg. produced a marked pressor response and symptoms of a typical attack in their patient. This same amount of mecholyl was given to normal patients, to hypertensive patients and to their initial patient with pheochromocytoma after removal of the tumor without producing a hyper-reactor response. This agent has since been used diagnostically with success in other proved cases of pheochromocytoma.[3,23,24] Guarneri and Evans came to the conclusion that mecholyl had a direct stimulating action on the chromaffin tumor. Although mecholyl exerts only slight nicotinic action,[26] this might be sufficient to stimulate secretion from such a large responding structure as a pheochromocytoma. The failure of curare to block such responses to mecholyl[22] cannot be regarded as a valid argument against this belief since the doses of curare used could not be expected to block at autonomic ganglia and the adrenal medulla.[25]

In an attempt to elucidate the mechanism of action of mecholyl in patients with pheochromocytoma a total of twenty-seven injections were given intra-arterially in six cats using the preparation of Feldberg and Minz. The doses varied from .08 to 50 μg. total. In only seven instances were there pressor responses (average 25 mm. Hg) usually preceded by a depressor phase (average 15 mm. Hg). On two occasions pressor responses were obtained without any prior depressor component. The responses of the blood pressure in this preparation to the injection were too variable to lend themselves to detailed study; however, since pure pressor responses were obtained on two occasions; it is suggested that mecholyl may cause epinephrine secretion by direct stimulation. The possibility of reflex stimulation of the adrenal medulla by a preliminary fall in blood pressure is ruled out when the response is purely pressor. Also, the adrenal glands in these preparations were denervated.

Whether or not this minimal nicotinic action is effective in causing epinephrine liberation from a pheochromocytoma is problematic. The marked pressor responses occurring in patients with this type of tumor as a result of the intravenous injection of mecholyl may be due in part to this nicotinic action and/or in part due to reflex stimulation from the fall in blood pressure. A direct stimulating action of mecholyl of the adrenal tumor tissue,[22] other than by possible nicotinic action, has not yet been proved.

Adrenergic Blocking Agents. Adrenergic blocking agents have recently been used as diagnostic aids in pheochromocytoma. Since liberation of epinephrine and/or nor-epinephrine from the tumor is responsible for the elevated arterial pressure in this disease, one would expect the adrenergic blocking agents to block this excitatory effect. On the other hand, if the hypertension were due to causes other than circulating sympathomimetic amines, one would expect these agents to have little effect. The adrenergic blocking agents were first suggested for use in cases of pheochromocytoma in 1941 by Biskind, Meyer and Beardner.[27] Six years later Goldenberg, Snyder and Aranow[28] reported the use of 933F as a new test for hypertension due to circulating epinephrine. They used this agent intravenously and in proved cases of pheochromocytoma a blood pressure fall was noted.[7,19,29–31] In normal patients or patients with essential hypertension 933F caused either no change of arterial pressure or a slight rise.[28,30] However, pressure rises occurring with the use of 933F can at times be quite marked, especially in patients with essential hypertension.[32] Recently a case of hypertensive encepha-

lopathy following the use of this drug was reported.[33]

In the original article describing the use of 933F[28] in pheochromocytoma (and also in a recent article[31]) the pressor responses are presumed to be due to stimulation of the central nervous system. Stimulation of the central nervous system by 933F in experimental animals has been described.[34] Pressor responses have been observed in anesthetized animals with the cord removed from C6 inferiorly along with bilateral vagotomy.[35] 933F will also cause hyperglycemia in experimental animals,[36] which is believed to be due to a central action mediated through the sympathoadrenal system.[37] By the use of cross circulation experiments in dogs Jourdan and Barrier[38] demonstrated that the benzodioxanes caused epinephrine secretion by direct action on the adrenal medulla and by reflex action. Although there is evidence to the contrary,[39] Nickerson[37] mentions that the benzodioxanes also exert a potent direct constrictor action on peripheral vessels. Therefore, there are several possible mechanisms for the marked pressor responses which can occur in certain patients when given the benzodioxane derivatives; stimulation of epinephrine liberation, central nervous system stimulation and direct peripheral vasoconstriction.

The marked pressor responses occurring in hypertensive patients are, however, probably not due to a primary medullary-stimulating action of 933F since one would expect this action to be in effect in those patients with adrenal medullary tumors. 933F is rarely followed by pressor responses in these patients.[44] Also, the intravenous use of dihydroergocornine (DHO-180) in a dose of 0.5 to 1.0 mg. intravenously has not been found to affect these pressor responses.[40] Because of the marked pressor responses which can occur in certain patients 933F is not an ideal drug for screening hypertensive patients to rule out pheochromocytoma.

The only other adrenergic blocking agents which have been reported to be of use in the diagnosis of pheochromocytoma are dibenamine (N, N, dibenzyl-beta-chlorethylamine hydrochloride), first reported by Spear and Griswold[41] in 1948, and C7337 (2 (N, para-tolyl-N [m-hydroxy phenol] aminoethyl)-imidazoline hydrochloride).[42] They probably exert blocking action by a specific block of some step in the process of excitation by sympathomimetic agents.[37] The value of dibenamine has been substantiated by other investigators[19,43] in the diagnosis of pheochromocytoma.

Dibenamine as well as benzodioxane derivatives has been found to have temporary therapeutic value in the care of patients with pheochromocytoma.[19] These agents have also been used preoperatively in the management of these cases.[19] Dibenamine should not be considered for use preoperatively since its use at this time precludes the later utilization of epinephrine as a pressor drug to combat the shock which occurs frequently during or immediately following the surgical removal of the tumor. It has been shown in experimental animals that blockade produced by dibenamine is ordinarily complete and responses to all doses of epinephrine are completely reversed.[37] However, the shorter acting adrenergic blocking agents such as the benzodioxane derivatives have been used preoperatively and may be of value. The blocking action of the benzodioxane derivatives is less complete than is the case with dibenamine and it can be overcome by using large doses of epinephrine.

In general, agents such as histamine, mecholyl and TEA, which cause an outpouring of epinephrine and/or nor-epinephrine from the adrenal medulla, are more useful than the blocking agents as diagnostic aids in the paroxysmal type of pheochromocytoma when the patient is normotensive between attacks. One can assume that other adrenal medullary-stimulating agents will be found of potential use in precipitating attacks. Other drugs which are now known to stimulate the adrenal medulla and cause epinephrine liberation are potassium, morphine, nicotine, tetramethylammonium and other related compounds. Since nicotine is a known stimulator of the adrenal medulla, one might suspect that patients with pheochromocytoma would respond differently to the stimulus of smoking as compared to normal or essential hypertensive patients.

SUMMARY

1. The pharmacologic basis of the various tests used in the diagnosis of pheochromocytoma is reviewed. These tests can be divided into two groups depending on whether a paroxysmal attack is induced or whether there is blockade of circulating epinephrine and/or nor-epinephrine.

2. Histamine and tetraethylammonium bromide probably induce attacks by a direct stimulating action on the adrenal medullary tissue.

3. The mechanism of action of mecholyl in inducing attacks in patients with pheochromocytoma is still not clear. It may be due in part to direct stimulation of the chromaffin tumor and/or reflex stimulation due to a preliminary fall in blood pressure.

4. The adrenergic blocking agents prevent the pressor responses to the circulating epinephrine and nor-epinephrine liberated from the adrenal tumors.

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Seminars on Arteriosclerosis

Protein-lipid Relationships in Human Plasma*

I. In Normal Individuals

ELLA M. RUSS, M.D., HOWARD A. EDER, M.D. *and* DAVID P. BARR, M.D.
with the technical assistance of Julie Raymunt and Ruth Aronson
New York, New York

It is currently believed that cholesterol, phospholipids and possibly other lipid fractions do not exist in the circulation in a free state but are combined with each other and with protein in the form of lipoproteins of large molecular weight. This is not a new concept. For fifty years it has been known that lipids cannot be completely extracted from plasma with ether.[1–5] In 1929 Macheboeuf[6] was able to isolate from serum substances which contained reproducible proportions of nitrogen and lipids and therefore could be appropriately called lipoproteins. Additional evidence of protein-lipid combinations has since been presented by Theorell,[7] Turner and Gibson,[8] Went and Goreczky,[9] and Blix, Tiselius and Svensson.[10]

More recently the protein fractionation methods of Cohn and his associates[11] have made possible the effective separation of alpha and beta lipoproteins and the demonstration that essentially all of the cholesterol of plasma forms a part of these two groups of proteins. Oncley's[12] analyses indicate that in pooled plasma approximately 25 per cent of the total cholesterol forms a part of the alpha lipoproteins and the remainder is found in the fraction containing the beta lipoproteins.

While these conditions have been preliminarily defined for pooled human plasma, it is to be expected that in individual and particularly in pathologic plasmas considerable variations might be encountered and that they might have significance in the diagnosis or prognosis of disease. The present study, which was initiated in 1949, had as its primary objective the exploration of such possible variations. Although this represented the chief focus of attention, the Cohn fractionation technic also permitted simultaneous observation of the distribution of other proteins with greater detail than had hitherto been possible with other methods.

This first report of protein-lipid relations represents a preliminary survey of the variations in a series of presumably normal individuals.

The subjects selected for study ranged from age eighteen to sixty-five and included both sexes. The great majority were medical students or members of the professional or non-professional staff of the hospital. Criteria for normality, always difficult to establish, consisted of vigorous participation in the work of the institution, the absence of any stigmas of acute or chronic disease and in most instances a recent health examination which had disclosed no abnormalities. A smaller number of the subjects were patients whose diagnostic surveys had revealed no deviation from robust health.

Chief emphasis was placed upon a group of twenty-four young men and twenty young women between the ages of eighteen and thirty-five, not only because they were sexually mature and gonadally active but also because they were statistically less liable to occult pathologic processes such as atherosclerosis and incipient cancer. Men and women between forty-five and sixty-five years of age were chosen with the hope of indicating effects of subsiding gonadal activity and aging.

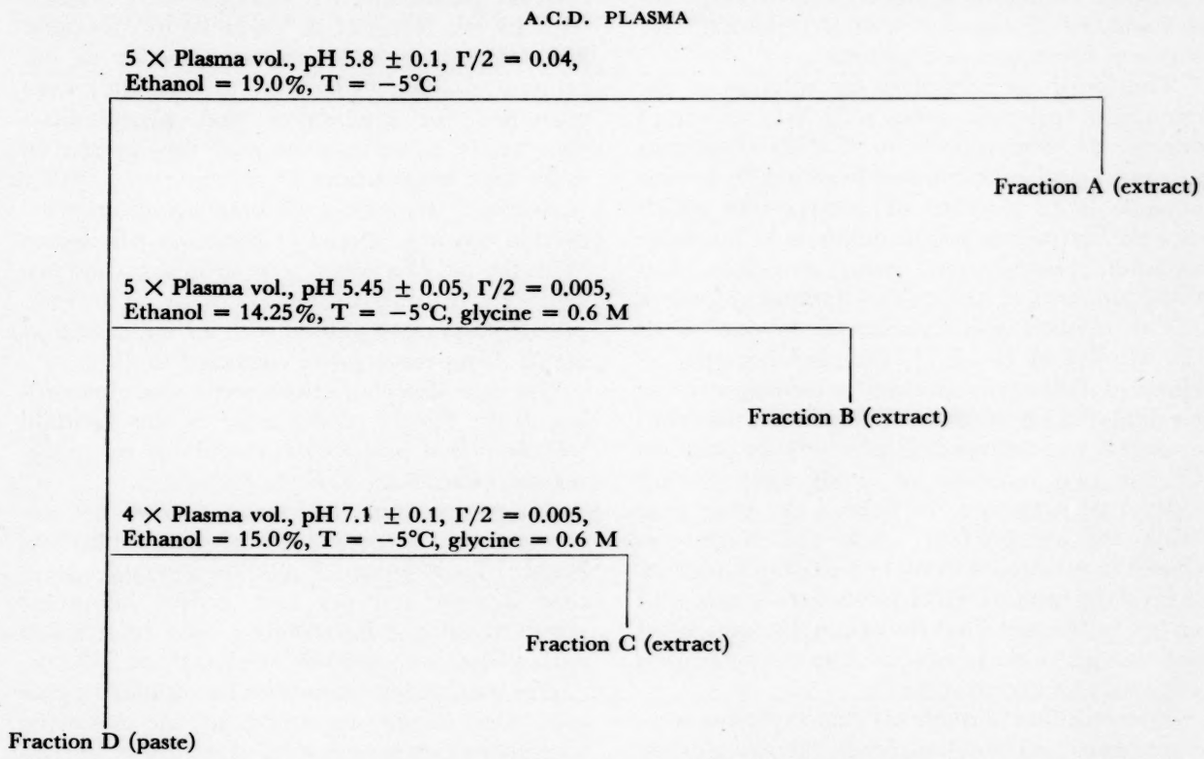
* From the Department of Medicine of the New York Hospital-Cornell Medical Center, New York, N. Y. The studies reported here have been aided by a grant from the U. S. Public Health Service and by support from the Commonwealth Fund and the Albert and Mary Lasker Foundation.

MATERIALS AND METHODS

Collections of blood specimens were obtained by venipuncture from individuals in the fasting state and were introduced at once into a graduated cylinder containing exactly 1.6 ml. acid citrate dextrose (A.C.D.)* solution for each 10.0 ml. blood; 25.0 ml. of blood yielded sufficient plasma for duplicate protein fractionations of 5.0 ml. aliquots and determinations of cholesterol, phospholipid and protein.

Fractionation of Proteins. Cohn's method 10 was employed.[11] This makes use of a system in which pH, ethanol concentration, ionic strength, protein concentration and temperature are critically controlled. For the separation of protein-protein complexes the dipolar ion glycine is introduced in high concentration.

The following shows the necessary conditions for the fixation of these variables whereby four major protein fractions are obtained, and also lists the proteins known to be extracted in each step of the method as we have carried it out:



Fraction D	Fraction C	Fraction B	Fraction A
Fibrinogen	β_1 Lipoprotein	γ Globulin	Albumin
Prothrombin	β_1 Lipid-poor euglobulins	(Traces albumin	α_1 Lipoprotein
Cold insoluble globulin	β_2 Globulins	and β_2 globulin)	α_1 Glycoprotein
Plasminogen	α_2 Globulin		α_2 Mucoprotein
β_1 Globulin	α_1 Globulin		α_2 Glycoprotein
(Small amounts α_2	(Traces fibrinogen and		β_1 Metal-combining globulin
and γ globulins)	albumin)		Small proteins and peptides

The first step of the procedure yields an extract representing Cohn's Fractions IV + V + VI and designated in this paper as Fraction A. It contains albumin, most or all of the alpha$_1$ lipoproteins, alpha$_1$ glycoprotein, alpha$_2$ mucoproteins, alpha$_2$ glycoproteins, beta$_1$ metal-combining protein and a considerable number of trace components, such as small proteins and peptides.

The second step yields an extract corresponding to Cohn's Fraction II and designated here as

* The A.C.D. solution contained per liter 22.0 gm. trisodium citrate (2.0 H_2O), 8.0 gm. citric acid monohydrate and 22.0 gm. dextrose.

Fraction B. It contains gamma globulins and little else. Traces of albumin, and $beta_2$ globulins and rarely $alpha_1$ and $beta_1$ globulins may be seen electrophoretically.

The third step yields an extract corresponding to Cohn's Fraction III_0 but here called Fraction C which contains $beta_1$ lipoproteins, $beta_1$ lipid-poor euglobulins, $alpha_2$ and $beta_2$ globulins with small amounts of $alpha_1$ globulin and traces of fibrinogen and albumin.

The fourth step involves the solution of the remaining proteins with 0.02 M trisodium citrate. It corresponds to Cohn's Fractions $I + III_{1,2,3}$ and is here called Fraction D. It contains a large number of components which include fibrinogen, prothrombin, cold insoluble globulin, plasminogen, $beta_1$ globulins, and small amounts of $alpha_2$ and gamma globulins.

The method was introduced to one of us (E. M. R.) in Dr. E. J. Cohn's laboratory at Harvard University during its development as an analytical procedure. The information thus obtained was followed as precisely as possible. Of the two technics for small scale protein separation suggested by Cohn[11] the procedure using the refrigerated angle centrifuge was chosen in preference to that employing filtration. It involved minimal risk from denaturation, with no loss in transfer from the original master tubes and was particularly adaptable to the separation and study of lipoproteins.

Five-milliliter aliquots of citrated plasma were fractionated within two hours following withdrawal of the blood, thereby minimizing protein changes due to enzymatic action, oxidation of lipoproteins and other deteriorations consequent to aging of plasma. The entire fractionation procedure was carried to completion within one working day with as little delay as possible between each step of the procedure.

In general, apparatus, reagents and separation procedures were those which had been described.[11] Modifications were as follows:

Fraction B was extracted with five instead of two plasma volumes of Reagent B* thus minimizing the occlusion of gamma globulins in the residual paste (Fraction C + D). The pH of the protein suspension following the addition of Reagent C* was adjusted to 7.1 to 7.2 with the addition of 0.03 to 0.04 ml. of sodium glycinate buffer.[13] This adjustment resulted in more complete separation of the $beta_1$ lipoproteins from the remaining D paste although small amounts of fibrinogen and prothrombin were extracted with the C fraction.

Following extraction of Fractions A and B the residual pastes in each instance were washed with 10 ml. Reagent A'* and 10 ml. Reagent B'* respectively. All washes were saved for protein analyses. From time to time they were examined for cholesterol and phospholipid content but in no instance were they present in more than slight traces.

Analytical Methods. pH determinations were carried out in a Model G Beckman pH meter. With the aid of a micro (one drop sample) glass electrode, measurements were made on 0.05 ml. protein suspension diluted with 0.2 ml. of 0.02 M NaCl. Temperature was corrected to 25°C.

Nitrogen determinations were made according to the Pregl[14] modification of the Kjeldahl technic. Total protein was calculated by applying the factor 6.25 × N.

Biuret determinations were carried out according to modifications[13] of the method of Mehl.[15] Each group of unknown samples were read against a 1 per cent bovine [albumin† standard using a Klett-Summerson colorimeter with a filter transmitting maximally at 540 mμ.

Dry weight determinations for estimating protein were made according to the following variation of the method of Armstrong and his associates:[16] The protein samples were dialyzed for five days at 2°C. against several changes of 0.3 M NaCl. Aliquots of the dialyzed material and final dialysate were dried to constant weight in an Abderhalden drying apparatus for two weeks at 40° to 50°C. P_2O_5 was used as a desiccant.

Cholesterol determinations were made on total plasma according to the method of Bloor, Pelkan and Allen[17] with modifications of Lever, Gurd, et al.[13] when applied to the protein fractions. Readings were made in a Klett-Summerson colorimeter with a filter transmitting maximally at 660 mμ.

Lipid phosphorus was determined by a modi-

* The reagents used to extract Fractions IV + V + VI, II and III_0 in Methods 10 were called Reagent A, Reagent B and Reagent C in Dr. Cohn's laboratory but not named as such in the publication.[11] It should be noted here that the composition of Reagent B in Methods 10 is not identical with Reagent B later reported in the publication of Lever and his associates.[13]

* Reagents A' and B' used here were those described by Lever and his associates.

† Purified bovine albumin was obtained from Armour and Company, Chicago, Ill.

fication of the method of Fiske and Subbarow.[18] A factor of 25 was used to convert lipid phosphorus to phospholipid. For plasma an aliquot of a 1to 25 Bloor extract with 3 to 1 alcohol-ether mixture was digested. For the protein fractions the method of Zilversmit et al.[19] was employed.

days at 2°c. against 1,000 ml. of the same Citrate Reagent which was changed daily. Dialysis of all four fractions was finally carried out in veronal citrate buffer, pH 8.6, ionic strength 0.1,[13] for two days with a change of buffer after twenty-four hours from 500 ml. to

TABLE I

PERCENTAGE AREA DISTRIBUTION OF PROTEIN COMPONENTS FROM ELECTROPHORETIC ANALYSIS OF PLASMA AND FOUR FRACTIONS

Specimen	Subject	Albumin (%)	α_1 (%)	α_2 (%)	β_1 (%)	β_2 (%)	ϕ (%)	γ (%)
Plasma	Sull.	51.5	8.9	9.1	14.6	6.4		9.5
	Lanm.	60.0	8.2	7.6	9.1	4.9		10.2
	Sand.	....	....	....	....	....		....
A(IV + V + VI)	Sull.	76.4	11.0	7.2	5.4	....	....	....
	Lanm.	....	....	....	....	....	....	....
	Sand.	86.9	6.8	2.8	3.5	....	....	....
B(II)	Sull.	1.4	....	....	....	4.1	....	94.5
	Lanm.	4.0	....	....	....	6.0	....	90.0
	Sand.	....	....	....	....	....	....	....
$C(III_0)$	Sull.	2.2	8.6	15.6	58.5	15.4		....
	Lanm.	....	....	19.8	60.5	19.8		....
	Sand.	....	3.2	16.0	52.4	11.7		16.7
$D(I + III_{1,2,3})$	Sull.	....	....	10.7	22.4	58.3		8.6
	Lanm.	....	....	13.8	32.2	54.0		....
	Sand.	....	....	....	....	....		....

An aliquot of the fraction was added to 10 per cent trichloracetic acid with stirring. After standing for ten minutes the tube was centrifugated. The supernatant was poured off and the tube allowed to drain. The precipitate was washed by resuspending it in trichloracetic acid, centrifugated and drained as before, and dissolved in dilute NaOH. A suitable aliquot was taken for digestion.

Electrophoretic analyses were performed occasionally to characterize the various fractions.

Concentration of extracts prior to electrophoresis was effected as follows: 1.0 ml. Zinc Reagent[13] was added respectively for each 12 ml. of Extract A and 19 ml. Extract C; 1.0 ml. of Sulfate Reagent[13] was added for every 1.0 ml. of Extract B. Following centrifugation supernatants were discarded and the resulting pastes, including the D paste, were redissolved with sufficient quantity of Citrate Reagent to yield a protein solution of 1.5 to 2.0 gm. per 100 ml.

Fractions A, B and C were then dialyzed two

1,000 ml. Total plasma was diluted to a concentration of approximately 2 per cent with the veronal citrate buffer and the dialysis procedure carried out the same way as for the individual fractions.

Electrophoresis was performed in an Aminco-Stern apparatus for two hours at 2°c. using a standard analytical cell.[20] With a current of 12 ma. a potential gradient of about 6.6 volts per cm. was maintained. The protein components were identified by their electrophoretic mobilities and their relative positions.

Calculations. In all calculations of protein, cholesterol and phospholipid on the original plasma and its fractions it was necessary to account for the dilution caused by the anticoagulant A.C.D. solution. All determinations were therefore multiplied by the factor 1.16.

Completeness of Separation of Proteins. The percentage area distribution of the protein components in plasma and the various fractions is indicated by electrophoretic determinations and

is illustrated in Table I. The calculations represent averages of the respective areas from both the ascending and descending limbs of all fractions except D. This fraction invariably started to clot before complete resolution was effected, resulting in poorer patterns of the descending than the ascending limb. Typical ascending patterns of normal plasma and Fractions A, B, C and D are shown in Figure 1. The alpha$_2$ and beta$_1$ peaks in the C fraction often migrated together with an increase in the mobility of the beta$_1$ peak.

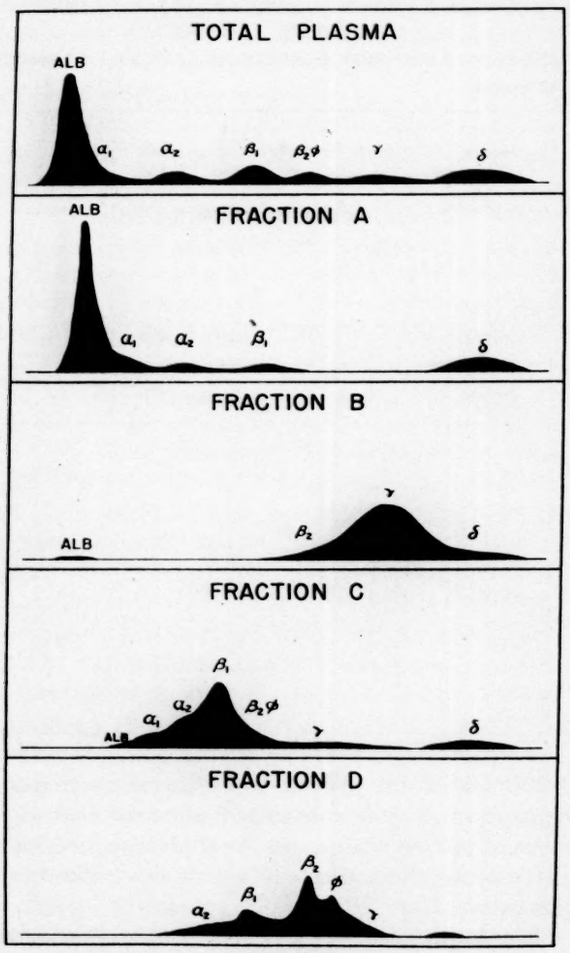


FIG. 1. Electrophoretic patterns (from ascending limb) of normal plasma and its four fractions.

Lever and his associates[13] obtained similar results. A condensation of their data is shown in Table II. The percentage area distributions are not completely comparable to our own because they measured only the proteins in Fraction IV + V while we estimated those in IV + V + VI (Fraction A), also because they combined Fractions I and III (Fractions C and D) while we measured III$_0$ (Fraction C) and I + III$_{1,2,3}$ (Fraction D) separately.

Nevertheless, it is apparent from the combined data that small amounts of albumin are carried into Fraction B (II) which also contains small amounts of beta$_2$ globulin; that Fraction C (III$_0$) usually contains small amounts of alpha$_1$ globulin; that variable but usually small amounts of gamma globulin appear in Fractions C and D, and that alpha$_2$ globulins may be present in considerable amounts in Fraction C.

Application of Analysis to Fractions. Biuret calibration curves were determined for purified bovine and human albumin,* Fraction I (Cohn) and Fraction II (Cohn). Dilutions of these proteins were made from a stock solution which had been prepared for dry weight determinations according to a method reported elsewhere in this publication. The photometric characteristics of these curves are shown in Figure 2. The unappreciable difference in slopes of the bovine and human albumin curves would indicate that bovine albumin may be used in place of human albumin as a standard in the biuret analyses.

* These fractions were supplied through the courtesy of Dr. E. J. Cohn.

TABLE II

ELECTROPHORETIC DATA CONDENSED FROM WORK OF LEVER AND HIS ASSOCIATES[13]

PERCENTAGE AREA DISTRIBUTION OF PROTEIN COMPONENTS

		Albumin (%)	α_1 (%)	α_2 (%)	β_1 (%)	β_2 (%)	ϕ (%)	γ (%)
Fraction IV + V	Average	81.3	7.8	4.7	5.5	0.7	....	.....
	Range	78–85	5–12	2–9	3–7	0.5–1.0	....	.....
Fraction II	Average	3.1	0.1	....	....	3.8	....	93.0
	Range	1–6	0–1	....	....	1–8	....	87–98
Fraction I + III	Average	0.5	3.5	13.3	49.4	26.5		6.8
	Range	1–2	1–6	8–29	30–61	18–36		2–11

Compared with the slopes of the albumin curves Fraction I deviates the least whereas the greatest deviation is exhibited by Fraction II. From Figure 2 it appears that every gram of human albumin is equivalent to 1.05 gm. Fraction I and 1.10 gm. Fraction II, and that every gram of bovine albumin is equivalent to 0.99 gm. Fraction I and 1.06 gm. Fraction II. Unfortunately, a biuret calibration curve for Fraction III_0 (Cohn) was not determined because a freshly purified sample was unobtainable.

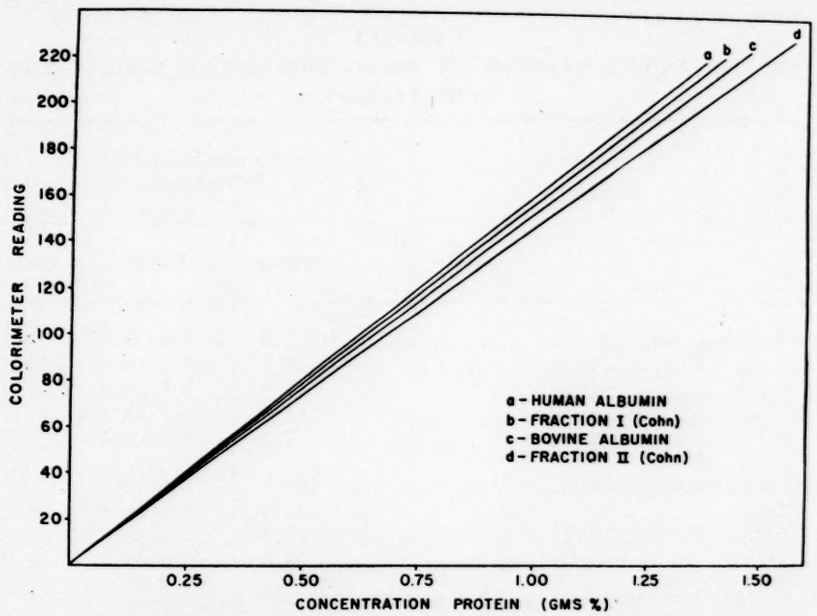


FIG. 2. Biuret calibration curves for purified human and bovine albumin, Fraction I (Cohn) and Fraction II (Cohn).

The protein concentrations of each fraction are recorded in this report as albumin equivalents for the following reasons: The application of corrections for each fraction based on biuret calibration curves (Fig. 2) would not be entirely valid since Fractions A, B, C and D are heterogeneous mixtures, the components of which may vary considerably in abnormal plasmas. Any correction of the biuret readings from estimations of the lipid content of lipoproteins would be of doubtful validity in plasma containing highly atypical lipoproteins. In general, the advantage of this method of presentation is that the biuret analyses may be compared directly with the Kjeldahl determinations of total protein. A disadvantage, however, lies in the difficulty of converting at once our weights and percentages of protein into equivalents obtained by dry weight determinations or those obtained by electrophoretic analyses.

Distribution of Cholesterol and Phospholipid in Fractions. Although most of the cholesterol and phospholipid were concentrated in Fractions A and C, small amounts were also found in Fractions B and D. The average per cent of cholesterol for B was 2.4, with a maximum of 8.6 and a minimum of 0 per cent; the average for D was 3.4, with a maximum of 12.2 and a minimum of 1.0 per cent. Usually less than 1 per cent of the total phospholipid was in Fraction B and between 2 and 3 per cent was in Fraction D, although occasionally larger proportions appeared in these fractions. The exact nature of the small amounts of lipoproteins in Fractions B and D is not known and because of the appreciable error in determining the small amounts of cholesterol and phospholipids their ratio has little validity in characterizing lipoproteins in these fractions. It is not unlikely that minute amounts of beta lipoproteins are extracted in complex with the gamma globulins. It is probable that small amounts of the beta lipoproteins are occluded in the D paste. Recently Green and Lewis[21] identified an $alpha_2$ lipoprotein in their ultracentrifugal studies which deserves attention here. Its position in the protein fractionation scheme shown previously is as yet unknown but the possibility that it may also contribute to the small amounts of cholesterol found in B and D cannot be excluded. Whether the small area of $alpha_1$ globulin occa-

sionally seen in the electrophoretic patterns of Fraction C is a lipoprotein cannot be stated at present.

Recovery of Protein, Cholesterol and Phospholipids. In comparing the protein analyses it was found that the sum of the biuret values in the fractions was almost always greater than the amount determined by the Kjeldahl method in the original plasma sample. The average recovery was 104 per cent with maximum of 109 and minimum of 97.

The sum of the cholesterol values from all fractions was somewhat less than the value determined by analysis of the original plasma. This may be partially explained in that the traces of cholesterol lost in the washes of A and B paste were too small to be determined accurately. The average recovery was 93.5 per cent with maximum of 103.0 and minimum of 86.0. Average phospholipid recovery from Fractions A and C amounted to 88 per cent of the amount determined on the original plasma with a maximum of 95 per cent and a minimum of 83 per cent.

TABLE III

DISTRIBUTION OF PROTEIN AND CHOLESTEROL IN YOUNG MEN AND WOMEN BETWEEN AGES EIGHTEEN TO THIRTY-FIVE

	Normal Women (20 Subjects)		Normal Men (24 Subjects)	
	Average	Range	Average	Range
Total protein, sum of fractions (gm. %)	7.3	6.8– 8.0	7.4	6.5– 8.5
Protein in A (%)	69.1	63.4–74.4	70.5	63.7–76.8
Protein in B (%)	11.9	9.1–14.9	11.4	8.0–15.3
Protein in C (%)	11.5	9.1–15.4	11.7	9.8–14.5
Protein in D (%)	7.4	4.2–12.2	6.5	4.9–10.9
Total cholesterol, sum of fractions (gm. %)	187	132–258	197	125–256
Cholesterol in A (%)	34.3	22.2–46.1	25.2	13.3–39.4
Cholesterol in B (%)	3.4	0– 8.4	2.9	0.9– 6.8
Cholesterol in C (%)	58.4	44.9–69.2	69.3	55.7–84.5
Cholesterol in D (%)	3.4	1.3–11.3	2.7	1.0– 7.2

RESULTS

Distribution of Protein and Cholesterol. Young Men and Women: Data on men and women between the ages of eighteen and thirty-five are shown in Table III. It will be seen that the average and in general the range of values for total protein, for the percentages of protein in each fraction and for total cholesterol, do not vary in the sexes. On the contrary, both the average and range of the percentage of cholesterol in Fraction A is higher and the percentage in Fraction C correspondingly lower, the average percentage being 34.3 ± 6.35 in young women and 25.2 ± 6.48 in young men. Statistically the difference is highly significant.

The difference between men and women is shown graphically in Figure 3 where the percentages of cholesterol in Fraction A are presented in relation to the total cholesterol of the plasma. A line is drawn arbitrarily at the level of 30 per cent. It will be seen that most of the men have values below and most of the women values above the line. It is also apparent that the percentage of cholesterol in Fraction A is not related to the level of total cholesterol.

Men and Women between the Ages of Forty-five and Sixty-five: In Table IV the observations on older men and women have been summarized. Differences between the sexes in the distribution of cholesterol are not apparent in this group. In both men and women the averages are slightly lower than those found in the younger men. Other significant findings not evident in the table should be stressed. In the group between eighteen and thirty-five the cholesterol in Fraction A was below 20 per cent in no women and only five men. In the older age group eleven of twenty-one men and nine of twenty women had values below 20 per cent. A tendency to reduc-

tion of the values for protein in Fraction A and an increase in proteins in Fraction C was apparent in several of the older men and women.

Variations in the Distribution of Protein and Cholesterol over Long Periods: Several of the normal women from the age group eighteen to thirty-five were tested frequently during the period of study. In Table v the protein and cholesterol values for the plasma and its fractions are presented for the different dates. The protein values fluctuated unpredictably within a relatively narrow range. Although the cholesterol values in the fractions also varied, it is worthy of note that individuals tended to maintain a relatively constant distribution. It is suspected that some of the variations may be related to the menstrual cycles. Further study of this phase is indicated.

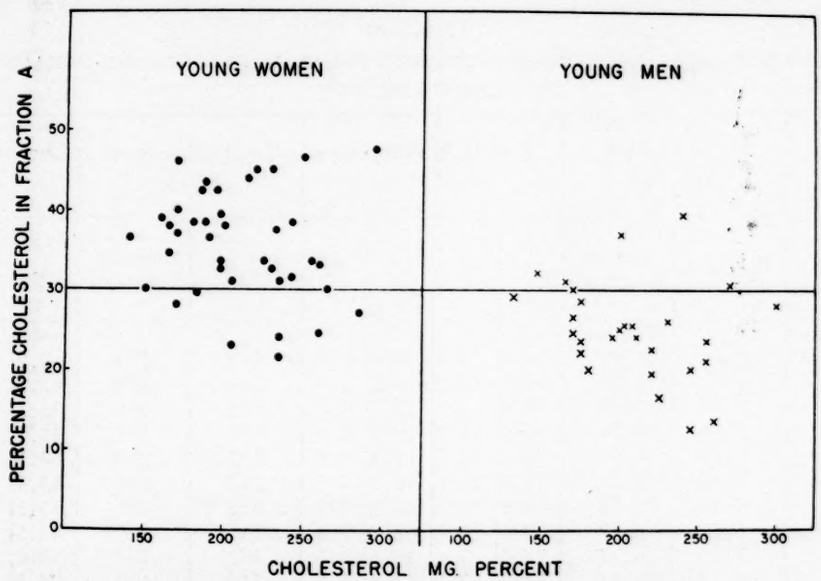


FIG. 3. Percentage cholesterol in Fraction A and total cholesterol of young men and women between ages eighteen to thirty-five.

TABLE IV

DISTRIBUTION OF PROTEIN AND CHOLESTEROL IN MEN AND WOMEN BETWEEN AGES FORTY-FIVE TO SIXTY-FIVE

	Normal Women (20 Subjects)		Normal Men (21 Subjects)	
	Average	Range	Average	Range
Total protein, sum of fractions (gm. %)	7.2	6.5– 8.5	7.3	6.6– 8.3
Protein in A (%)	63.7	57.7–67.3	67.5	59.7–73.2
Protein in B (%)	13.8	11.1–18.9	12.5	7.7–15.9
Protein in C (%)	13.3	9.5–17.8	12.4	9.6–15.0
Protein in D (%)	9.2	5.9–11.9	7.6	4.7–12.8
Total cholesterol, sum of fractions (mg. %)	252	163–340	239	167–268
Cholesterol in A (%)	23.4	11.7–38.4	22.9	12.4–40.7
Cholesterol in B (%)	1.5	0– 3.8	1.8	0– 8.6
Cholesterol in C (%)	71.5	52.4–81.1	71.2	54.2–83.3
Cholesterol in D (%)	3.5	1.0– 7.3	4.1	1.8–12.2

Distribution of Phospholipid in the Plasma and the Relationship of Cholesterol to Phospholipid. In

Table VI weights and percentages of phospholipid are listed and comparison is made between the cholesterol-phospholipid ratios of plasma, Fraction A and Fraction C.

The cholesterol-phospholipid ratio by weight for plasma averaged 0.93 with variations from 0.75 to 1.14. In Fraction A the average was 0.52 with relatively small variations from 0.44 to 0.62. For Fraction C the mean ratio was 1.34 but the variations were considerably larger and ranged from 1.10 to 1.53. These values agree in general with the ratios determined for Fractions A and C by Oncley[12] and Lever.[13] Our somewhat higher values in Fraction C may possibly be attributed to differences in method of analysis.

The fairly constant and considerable differences between the cholesterol-phospholipid ratios in Fractions A and C suggest that lipoproteins of differing chemical constitution are separated in the two fractions. The constancy of these ratios in a variety of plasmas is contributory evidence of the validity of the separation of the two lipoproteins.

TABLE V

VARIATION IN DISTRIBUTION OF PROTEIN AND CHOLESTEROL IN YOUNG NORMAL FEMALE SUBJECTS OVER LONG PERIODS

Date	Subject	Total Protein (gm. %)		Protein in Fractions (%)				Total Cholesterol (mg. %)		Cholesterol in Fractions (%)			
		Kjeldahl	Sum of Fractions	A	B	C	D	Plasma	Sum of Fractions	A	B	C	D
1- 6-50	Carl.	6.96	6.66	74.0	8.9	8.8	8.3	252	210	46.2	5.3	43.8	4.2
1-19-50	Carl.	6.92	6.87	73.4	10.2	9.5	6.9	185	187	42.3	6.0	49.6	2.1
2- 3-50	Carl.	7.39	7.41	71.2	11.4	10.7	6.6	253	246	33.5	7.5	55.7	2.8
2- 1-51	Carl.	7.50	7.74	69.3	11.7	10.9	8.1	226	192	44.1	0.8	50.8	4.4
3- 1-51	Carl.	7.10	7.59	71.6	10.9	9.4	8.1	242	212	38.5	3.7	52.0	5.4
3- 9-51	Carl.	7.10	7.77	71.9	11.0	10.3	6.8	232	216	45.4	4.0	46.7	4.1
3-21-51	Carl.	7.00	7.15	71.5	10.3	10.5	7.7	222	214	45.3	1.4	49.2	4.0
1-12-50	Russ.	6.65	6.52	77.0	8.0	8.0	7.0	191	185	42.5	3.0	51.0	3.5
1-24-50	Russ.	7.05	6.77	77.1	7.4	10.6	4.9	172	183	40.0	0.2	58.0	1.8
2- 3-50	Russ.	6.67	6.47	75.3	7.3	11.8	5.6	163	162	34.3	3.3	60.3	2.1
5- 8-50	Russ.	6.88	7.11	73.4	11.4	11.7	3.5	181	163	38.3	3.9	57.0	0.8
5-10-51	Russ.	6.80	6.96	74.0	9.5	16.5		177	156	38.2	0.9	60.9	
12-28-49	Cand.	7.00	6.40	69.7	12.4	9.4	8.5	167	158	33.8	1.1	58.9	8.2
1-19-50	Cand.	7.18	6.83	71.7	11.9	9.3	7.1	183	171	29.6	5.5	61.9	3.0
2- 3-50	Cand.	6.95	7.23	70.7	12.5	10.0	6.8	204	196	31.3	8.0	59.0	1.7

COMMENTS

Selection of Normal Subjects. In a study of dynamic factors such as proteins and lipids, the selection of normal individuals presents many difficulties which greatly increase when men and women in the later decades of life are considered. Opinions concerning freedom from disease are relative. Furthermore, it is not possible at present to evaluate the influence of a great variety of incidents such as the menstrual cycle, fatigue, dietary excesses on days preceding tests, gastrointestinal disturbances and subclinical infections. Heredity, body build and constitution are also factors which could modify the protein and lipid metabolism and distribution. The recent work of Gertler, Garn and Sprague[22] clearly indicates that the levels of total cholesterol in the plasma are higher in mesomorphic individuals. The differences which are apparent in our series between young men and young women indicate a possible effect of hormonal substances and emphasize the question of the influence of the menstrual cycle with its wide swings in pituitary, adrenal and gonadal activity. Febrile infections are known to modify the level of cholesterol[23–25] in the circulating blood, and some of them undoubtedly affect the distribution of protein substances. Establishment of truly normal levels could be made only on the basis of an enormous number of observations. Our own determinations in this paper

must be regarded as indicative rather than decisive.

Influence of Age. Our limited data indicate that the group of individuals between the ages of forty-five and sixty-five contains a greater number of persons whose percentage of cholesterol in Fraction A falls in the lower ranges. Several observations, however, show clearly that high values may be obtained in individuals of the older group. The question whether the lower values are the effect of age or of hidden disease cannot be answered at the present time.

Influence of Sex. The influence of sex on the distribution of lipids, which is perhaps the most significant contribution of this study, has already been indicated. That it is dependent upon endocrine factors is suggested by the differences between young men and women, and their absence in the older age groups. It is a factor which must be considered in the investigation

TABLE VI

PHOSPHOLIPIDS AND CHOLESTEROL-PHOSPHOLIPID RATIOS IN PLASMA AND FRACTIONS

Subject	Age	Sex	Total Plasma		Fraction A		Fraction C	
			mg. %	Chol./P'lipid.	mg. %	Chol./P'lipid.	mg. %	Chol./P'lipid.
Part	25	M	201	1.00	82	.52	86	1.40
Sand	25	M	189	.95	83	.48	88	1.40
Week	22	F	188	1.05	104	.56	75	1.44
Wilk	22	F	219	.85	121	.57	63	1.30
Less	25	F	232	.89	96	.46	99	1.22
Carl	28	F	246	.92	138	.61	67	1.45
Raym	25	F	301	.76	141	.50	129	1.10
Brei	22	F	212	.75	112	.45	74	1.18
Russ	34	F	212	.84	111	.54	79	1.20
Morg	28	F	290	.91	126	.56	115	1.42
Hiza	22	F	177	.97	95	.57	61	1.30
Wend	22	F	216	.80	129	.55	57	1.22
Fisc	22	F	216	.95	114	.48	87	1.53
Musc	46	M	230	1.01	92	.42	123	1.34
Will	46	M	222	1.04	99	.55	90	1.50
Wrig	50	M	287	1.07	91	.50	162	1.38
Barr	60	M	305	.80	142	.46	125	1.19
Rezn	56	M	275	.95	90	.50	141	1.26
Simo	45	M	246	.93	129	.62	92	1.49
Dega	49	M	293	.99	94	.54	162	1.28
Gree	48	M	282	.94	88	.50	174	1.23
Stev	49	M	283	1.14	78	.53	180	1.44
Wolf	53	M	218	.78	130	.52	71	1.27
Oppe	48	M	217	.82	118	.50	82	1.29
Tols	54	M	290	.85	100	.48	141	1.42
Catt	59	M	278	.93	128	.48	120	1.34
Smil	63	M	284	.92	140	.53	119	1.29
Bric	54	M	235	1.13	76	.44	120	1.53
Nuss	56	F	275	1.09	100	.50	145	1.49
Rouc	59	F	277	.94	97	.49	137	1.45
Doyl	51	F	244	.88	104	.53	111	1.22
Deko	60	F	322	1.08	100	.51	185	1.37
Wall	61	F	327	1.08	91	.44	189	1.44
Erpf	58	F	311	1.03	121	.46	164	1.46
Hanl	58	F	274	.91	130	.50	113	1.43
Rehd	56	F	241	.93	118	.61	83	1.40
Ivos	62	F	294	.99	87	.47	173	1.28
Barr	59	F	212	1.04	87	.49	109	1.43
Mean	..	..	253	.95	107	.51	116	1.35
Range	..	..	188–327	.75–1.14	76–142	.42–.62	61–189	1.10–1.53

of pathologic conditions and which merits special studies of the influence of the menstrual cycle, castration, the menopause and the effects of hormonal agents in therapy.

Cholesterol-Phospholipid Ratios. Recently interest has been revived in the observation of Boyd[26] that phospholipids play an important role in maintaining the stability of plasma and lipid emulsions. Considerable data have accumulated as to the relative concentrations of cholesterol and phospholipid in plasma and the significance of this ratio in various disease states.[27–29] Our own data are of interest because they permit clearer definition of the factors influencing the ratio in plasma. It is apparent that this is a function of at least three variables: (1) the ratio of cholesterol and phospholipid in Fraction A, (2) the ratio in Fraction C and (3) the distribution of the total lipid between the two fractions. Since the absolute variation of the ratios in Fraction A is slight, this factor is of relatively minor importance. In Fraction C the variation of the ratios is greater to the extent that it has considerable weight in determining the plasma ratio. When a large portion of the plasma lipid is present in Fraction A with its low cholesterol-phospholipid ratio, the ratio of the unfractionated plasma will tend to be low. With a preponderance of lipid in Fraction C the plasma ratio will tend to be high. This trend, however, is not always evident in the ratios of unfractionated plasma because of the variability of the ratios in each of the fractions.

SUMMARY AND CONCLUSIONS

1. The Cohn microfractionation method No. 10 for the separation of proteins has been applied to the plasma of eighty-five presumably normal individuals ranging in age from eighteen to sixty-five. Protein, cholesterol and phospholipids have been determined upon the original plasma and upon each of the fractions.

2. Confirmatory evidence has been offered to support the thesis that essentially all of the cholesterol and phospholipid of plasma is combined with protein in the form either of alpha lipoproteins or beta lipoproteins.

3. The cholesterol-phospholipid ratio of the fraction of plasma containing the alpha lipoproteins averages 0.5 while that containing the beta lipoproteins has an average ratio more than twice as great. Considerable variation in the ratios of both fractions is apparent, the range for alpha lipoproteins being 0.42 to 0.62 and for beta lipoproteins 1.10 to 1.53.

4. Plasma of young women between the ages of eighteen and thirty-five contains more alpha lipoprotein and less beta lipoprotein than that of men of comparable age. Otherwise there is no significant difference in the protein lipid distribution of the sexes.

5. Presumably normal men and women between the ages of forty-five and sixty-five do not exhibit this sex difference in lipid distribution. A considerable number of them, both men and women, reveal a tendency to relatively low values for albumin and alpha lipoprotein and relatively large concentrations of beta lipoprotein.

Acknowledgment: We wish to express our appreciation to Dr. Edwin J. Cohn, Dr. F. R. N. Gurd and Dr. J. L. Oncley for the opportunity to acquire the technic of Methods 10 in the University Laboratory of Physical Chemistry as related to Medicine and Public Health, Harvard Medical School, and for all the invaluable advice and assistance pertaining to the fractionation method and electrophoresis.

We gratefully acknowledge the analytical determinations performed by Miss Hanna Fischer and Miss Mary Donahue.

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Protein-lipid Relationships in Human Plasma*

II. In Atherosclerosis and Related Conditions

DAVID P. BARR, M.D., ELLA M. RUSS, B.S. *and* HOWARD A. EDER, M.D.
with the technical assistance of Julie Raymunt and Ruth Aronson
New York, New York

In a previous communication[1] we reported results of studies with the Cohn protein microfractionation method 10[2] on the plasma of presumably normal men and women. It appeared that essentially all of the cholesterol and phospholipid of normal plasma is combined with protein and presumably in combination either with alpha lipoproteins or with beta lipoproteins. The fraction of the plasma containing alpha lipoproteins had a cholesterol-phospholipid ratio averaging about 0.5 by weight whereas the fraction in which beta lipoproteins were concentrated exhibited ratios over twice as great. In comparing the sexes no significant differences were encountered except that the plasma of young women contained more alpha lipoprotein and consequently less beta lipoprotein than that of young men. In the age period between forty-five and sixty-five this sex difference was no longer detectable but it was noted that relatively low values of alpha lipoprotein were encountered more frequently in the older men and women.

Differences thus encountered in the sexes and with aging led to the inference that the distribution of lipids between the two groups of lipoproteins might be relevant to the genesis of atherosclerosis. This investigation was therefore undertaken to determine whether there are significant abnormalities of lipid distribution in the plasma of patients with atherosclerosis or with conditions that are known to predispose to its development.

The present report is concerned with single or multiple studies of thirty-three patients who were known or thought to be suffering from complications of atherosclerosis, two who exhibited advanced lesions of familial xanthoma tendinosum with hypercholesterolemia, thirty-five diabetics and twelve nephrotics.

In the atheromatous group selection of patients was subject to the usual difficulties in diagnosis and necessitated the adoption of arbitrary criteria. Twenty-eight of the subjects had survived one or more myocardial infarctions; three presented clinical and electrocardiographic signs of progressive coronary insufficiency; one had arteriosclerotic gangrene of the foot; and the other was shown by surgery to have extensive atheroma with completely obstructing thrombus of the descending aorta. The group included twenty-nine men and four women varying in age from twenty-eight to sixty-six.

Of the diabetics seventeen were women and eighteen were men. Their age ranged from four to eighty-four, and nineteen were below the age of thirty-six. They varied greatly in the severity of their disease, the duration of the diabetic state, the incidence of ketotic and hypoglycemic attacks and the clinical evidence of vascular complications.

The nephrotic group was equally divided between the sexes and included subjects ranging from one to fifty-five. All of them were suffering from edema and other active manifestations of their disease at the time their study was undertaken.

The normal subjects with whom these pathologic cases are to be compared were those reported in the previous communication. They were normal in the sense that they displayed

* From the Department of Medicine of the New York Hospital-Cornell Medical Center, New York, N. Y. The studies reported here have been aided by a grant from the U. S. Public Health Service and by support from the Commonwealth Fund and the Albert and Mary Lasker Foundation.

no clinically evident defect or illness. The possibility that some of them were atheromatous cannot be excluded.

MATERIALS AND METHODS

In a previous communication[1] the procedures and methods used in this study were described, with some discussion of their limitations and their applicability to individual samples of normal plasma. In the study of abnormal conditions no variations in technic have been introduced. As in the previous paper, Cohn's fraction IV + V + VI is designated here as Fraction A; his Fraction II as Fraction B; his Fraction III_0 as Fraction C; and the remaining paste of I + $III_{1,2,3}$ as Fraction D.

In all but nephrotic plasmas the pH of the protein suspensions at each step of the fractionation procedure was within the indicated range.[1] In the nephrotics, however, the pH readings of suspensions A + B + C + D and B + C + D were in general 0.2 to 0.3 unit lower than the anticipated range. For this reason it is quite probable that the separation of the protein components in nephrotic plasmas was not as complete. The necessity of further experimentation with the extraction reagents is indicated.

TABLE I

PERCENTAGE AREA DISTRIBUTION OF PROTEIN COMPONENTS FROM ELECTROPHORETIC ANALYSIS

Specimen	Subject	Diagnosis	Albumin (%)	α_1 (%)	α_2 (%)	β_1 (%)	β_2 (%)	ϕ (%)	γ (%)
Plasma	Sull	Normal	51.5	8.9	9.1	14.6	6.4		9.5
	Weid	Atherosclerosis	47.7	10.6	11.1	15.5	6.7		8.3
	Bart	Diabetes	44.4	9.4	11.8	13.7	6.9		13.9
	Geff	Xanthoma	44.0	9.0	12.1	16.4	7.7		10.3
	Malo	Nephrosis	23.9	9.2	23.1	19.1	15.9		8.8
A(IV + V + VI)	Sull	Normal	76.4	11.0	7.2	5.4	....		....
	Weid	Atherosclerosis	72.4	14.4	8.4	4.9	....		....
	Bart	Diabetes	75.0	10.4	7.8	6.4	....		....
	Geff	Xanthoma	74.6	11.0	8.9	5.6	....		....
	Malo	Nephrosis	52.3	13.9	24.7	9.1	....		....
B(II)	Sull	Normal	1.4	....	....	....	4.1	....	94.5
	Weid	Atherosclerosis	....	....	....	....	....	....	....
	Bart	Diabetes	1.0	....	....	....	3.0	....	96.0
	Geff	Xanthoma	....	....	....	....	....	....	....
	Malo	Nephrosis	....	....	....	....	....	....	....
$C(III_0)$	Sull	Normal	2.2	8.6	15.6	58.5	15.4		....
	Weid	Atherosclerosis	....	....	11.7	64.9	23.5		....
	Bart	Diabetes	....	10.6	24.7	48.4	16.0		....
	Geff	Xanthoma	....	....	27.0	54.6	18.5		....
	Malo	Nephrosis	....	....	22.0	54.5	23.5		....
D(I + $III_{1,2,3}$)	Sull	Normal	....	....	10.7	22.4	58.3		8.6
	Weid	Atherosclerosis	....	....	....	10.7	52.8		16.2
	Bart	Diabetes	....	....	....	12.3	68.5		19.3
	Geff	Xanthoma	....	....	....	12.2	71.6		16.2
	Malo	Nephrosis	....	....	....	8.8	66.6		24.5

In Table I comparison is made between the electrophoretic values for percentage area distribution of the protein components in normal and abnormal plasmas and their fractions. Essentially the same limitations as in normal blood are evident. All but a small fraction of $alpha_1$ globulin is concentrated in Fraction A and most of the $beta_1$ globulin in Fraction C.

It appears that in xanthoma, diabetes and nephrosis the recovery in Fraction A of albumin and $alpha_2$ globulin may not be as complete as in normals. In one diabetic the percentage of $alpha_1$ globulin in Fraction C is somewhat

TABLE II

DISTRIBUTION OF PROTEIN IN PLASMA AND FRACTIONS

Subject	Age	Sex	Total Protein (gm. %)		Protein in Fractions (%)				Total Cholesterol (mg. %)		Cholesterol in Fractions (%)				Remarks
			Kjeldahl	Sum of Fractions	A	B	C	D	Plasma	Sum of Fractions	A	B	C	D	
Atherosclerosis															
Dele	59	M	6.8	7.0	57.8	12.8	11.0	18.4	182	179	5.6	3.5	78.0	13.3	Myocardial infarction 29 days before observation
Krau	42	M	6.5	7.1	63.9	12.3	23.8		361	338	6.8	0.7	92.5		Myocardial infarction 11 wk. before
Ston	66	M	...	8.3	45.1	21.8	13.4	19.2	253	241	6.9	0.0	75.9	16.8	Myocardial infarction 6 wk. before
Curt	32	M	6.9	7.0	62.4	11.1	15.3	11.1	291	273	7.0	8.7	81.1	3.1	Myocardial infarction 9 days before
Giac	61	M	7.3	7.3	59.3	14.8	15.8	10.1	394	452	8.3	5.1	84.5	2.1	Myocardial infarction 30 days before
Newm	45	M	...	7.9	58.5	11.6	15.8	14.0	318	291	8.6	1.0	82.1	8.4	Myocardial infarction 3 wk. before; glycosuria following coronary occlusion
Rals	49	M	6.6	6.7	64.3	9.1	17.0	9.6	314	292	8.7	2.3	86.4	2.7	Myocardial infarction 4 days before
Saks	38	M	7.4	7.6	63.7	14.2	13.4	8.7	330	328	9.4	1.6	84.8	4.2	Angina pectoris for 8 yr.; EKG changes of coronary insufficiency; albuminuria
McDo	48	M	7.4	8.0	56.6	15.1	16.4	11.8	380	349	9.4	1.0	83.7	6.1	Myocardial infarction 2 mo. before
Harr	39	M	7.8	8.0	55.4	14.4	14.0	16.2	253	223	9.5	0.0	78.1	12.6	Myocardial infarction 9 mo. before; bundle branch block
Shar	44	M	6.4	7.1	44.4	15.7	24.1	15.8	310	290	9.9	2.0	83.6	3.0	Repeated myocardial infarctions for 6 yr.; observation 10 days before death
Laro	37	M	7.3	7.9	68.4	12.3	12.4	7.2	266	251	10.3	0.5	85.6	3.5	Myocardial infarction at 35; familial muscular atrophy
Newc	32	M	7.4	7.5	59.5	11.3	16.3	12.8	392	368	10.3	1.6	85.0	3.1	Myocardial infarction 7 wk. before
Aron	48	M	7.1	6.4	55.7	18.9	17.2	8.2	296	329	11.2	4.7	82.8	1.4	Atherosclerotic gangrene of foot
Borz	48	M	6.8	7.1	67.1	8.9	14.3	9.7	237	208	12.7	1.7	82.8	2.9	Atherosclerotic occlusion of abdominal aorta and iliac artery
John	44	M	7.0	7.2	64.7	10.4	18.3	6.6	264	230	12.9	3.3	81.5	2.4	Myocardial infarctions at 39 and 43
Lurr	36	M	6.9	6.9	64.0	10.0	15.4	10.6	294	266	13.3	0.0	84.2	2.4	Myocardial infarction 17 days before; transient glycosuria
Loga	37	M	6.7	7.1	69.6	8.8	21.5		....	242	13.8	0.9	85.5		Myocardial infarction 75 days before
Weid	42	M	7.1	7.2	65.7	11.0	15.0	8.4	298	267	14.5	1.6	81.4	2.4	Precordial pain for 10 yr.; peptic ulcer
Duff	64	F	...	7.5	55.2	15.5	12.4	16.8	264	254	14.6	0.8	73.3	11.4	Myocardial infarction 14 days before
Nard	46	M	7.4	7.9	62.4	17.5	10.9	9.4	258	243	14.9	2.0	78.2	4.8	Myocardial infarction at 38
Geno	40	M	7.3	7.8	59.6	18.3	13.3	8.8	226	199	15.7	2.3	78.9	3.1	Myocardial infarction at 35 and 38
Sele	28	M	7.0	7.1	66.7	11.9	13.3	8.1	355	330	16.3	3.4	78.4	2.1	Myocardial infarction, diagnosed by EKG; congestive heart failure
Rodm	64	M	6.6	6.1	56.3	8.5	19.1	16.1	206	197	16.5	5.9	74.7	2.8	Myocardial infarction 3 days before
Cass	36	M	6.8	7.1	66.2	12.8	14.1	6.9	204	175	17.0	2.1	80.0	1.9	Myocardial infarction at 33
Schu	36	M	6.9	7.1	66.0	11.2	13.5	9.2	286	271	17.0	3.1	75.4	3.7	Myocardial infarction 9 months before
Wein	44	M	6.8	7.2	64.4	14.2	14.9	6.5	266	230	17.2	1.1	81.0	0.9	Progressive coronary insufficiency—peripheral arteriosclerosis
Brow	52	M	7.8	8.0	64.5	12.3	14.9	8.3	350	317	17.4	0.0	80.5	2.0	Myocardial infarction 5 wk. before
Sulp	53	F	7.4	7.5	51.9	17.1	15.7	15.3	269	258	19.9	0.0	69.7	10.3	Myocardial infarction 3 wk. before
Mysz	57	F	6.8	6.7	56.4	13.5	12.6	17.4	159	155	20.0	2.7	73.0	4.3	Myocardial infarction 8 days before; angina pectoris for 5 yr.
Ober	41	M	6.6	6.9	66.7	10.3	14.8	8.2	214	205	23.1	1.1	72.3	3.5	Myocardial infarction at 40
Marr	42	F	7.5	7.8	61.1	17.2	10.1	11.6	197	177	23.2	4.1	61.0	11.8	Myocardial infarction at 40
Bruc	52	M	6.8	6.7	57.2	12.3	12.7	17.8	149	135	25.6	4.9	63.8	5.8	Myocardial infarction 9 days before; subtotal gastrectomy 6 mo. before
Xanthoma Tendinosum															
Geff	44	M	7.7	8.1	69.6	9.1	13 8	7.4	384	337	5.6	0.0	91.2	3.1	Xanthoma of tendon sheaths; myocardial infarction
Book	50	M	7.4	7.9	63.3	12.2	15.1	9.3	579	508	6.2	2.2	89.3	2.5	Xanthoma of tendon sheaths; angina pectoris; peripheral arteriosclerosis
Diabetes															
Gust	24	F	7.0	7.0	37.7	9.7	38.7	13.7	860	794	5.6	0.2	91.4	3.1	Diabetic at 10; Kimmelstiel-Wilson syndrome; retinitis proliferans; autopsy: extensive atheroma
Ship	25	M	7.3	7.4	53.6	9.8	36.5		291	280	6.8	0.0	93.2		Diabetic at 4; no vascular complications; nonspecific pericarditis
Adle	69	M	6.5	6.3	64.7	12.8	15.1	7.4	203	176	7.8	0.0	90.9	1.0	Observation during diabetic coma and mesenteric thrombosis
Seid	43	M	8.3	8.7	63.6	12.3	14.0	10.1	325	294	8.0	0.0	84.9	7.2	Diabetic at 24; retinitis; albuminuria
Smit	42	M	7.0	7.5	61.5	9.8	17.4	11.3	820	705	8.3	0.2	87.9	3.5	Observation 6 days after diabetic coma with excessive hyperlipemia
Mart	21	F	6.1	6.6	57.2	10.7	23.8	8.1	390	360	9.6	0.9	87.7	1.8	Diabetic at 10; occasional albuminuria; necrobiosis lipoidica
Wilm	60	F	7.4	7.8	58.1	12.6	15.9	13.8	346	346	10.2	0.6	85.8	3.5	Diabetic at 52; hypertension; pulmonary tuberculosis
Zubc	84	F	6.3	6.5	58.9	10.8	18.7	11.6	196	193	12.5	1.2	84.0	2.4	Mild diabetes of unknown duration
Webb	64	F	7.3	7.8	53.8	13.4	20.7	12.1	268	253	13.5	0.0	82.5	3.8	Diabetic at 63; myocardial infarction 18 days before observation
Bart	16	F	7.2	7.5	64.3	11.4	17.2	7.1	290	244	13.6	0.9	84.0	1.6	Diabetic at 13; no vascular complications
Benj	23	M	6.6	6.6	68.8	5.3	19.5	6.4	237	216	13.6	0.4	84.1	1.9	Diabetic at 16; retinitis proliferans; albuminuria
Wahr	72	M	7.3	7.1	62.6	13.2	16.5	7.6	319	294	14.8	0.6	82.3	2.4	Mild diabetes discovered at age of 70; no vascular complications

TABLE II—(*Continued*)

Subject	Age	Sex	Total Protein (gm. %)		Protein in Fractions (%)				Total Cholesterol (mg. %)		Cholesterol in Fractions (%)				Remarks
			Kjeldahl	Sum of Fractions	A	B	C	D	Plasma	Sum of Fractions	A	B	C	D	
Flin.....	52	F	6.6	7.0	60.7	15.7	15.3	8.2	272	262	15.8	1.2	79.2	3.7	Mild diabetes discovered at age of 50; no vascular complications
Stew.....	4	F	7.1	7.7	67.4	9.7	16.6	6.3	276	272	16.0	1.5	81.4	1.3	Glycosuria discovered 2 mo. before observation
Faja.....	59	M	6.7	7.0	66.2	11.6	16.5	5.7	186	179	17.6	1.0	79.8	1.3	Mild diabetes of unknown duration; retinitis proliferans
Bala.....	17	M	6.7	6.5	60.9	12.0	14.9	12.2	181	163	18.0	1.5	77.1	3.5	Diabetic at 14; no vascular complications
Mosc.....	22	F	8.0	8.2	57.3	18.8	16.1	7.8	208	182	18.2	2.1	79.0	1.0	Diabetic at 14; intermittent albuminuria; retinitis proliferans
Grif.....	25	F	5.8	6.9	57.7	10.8	23.2	8.4	196	166	19.0	0.5	76.3	4.3	Mild diabetes for 7 yr.; coma 3 days before observation
Pola.....	30	M	7.1	7.3	67.5	10.9	14.6	7.1	201	185	19.9	4.1	73.2	2.8	Diabetic at 15; high insulin requirement; healed tuberculosis
Capp.....	35	F	7.1	7.3	59.5	17.6	13.4	9.3	210	202	20.4	4.0	71.1	4.5	Obese; mild diabetes unknown duration; no vascular complications
Stei.....	34	F	6.2	7.0	70.7	9.1	13.0	7.5	262	251	21.2	1.1	76.7	1.1	Mild diabetes duration unknown; no vascular complications
Henr.....	22	F	6.4	6.8	67.4	11.3	16.7	4.6	171	172	21.3	2.8	74.6	1.3	Diabetic at 9; no vascular complications
Graf.....	40	M	7.0	7.2	71.5	11.5	9.3	7.8	208	189	22.0	8.0	67.9	2.0	Very mild diabetic, accident prone, neurotic; no vascular complications
Wrab.....	41	M	6.6	6.9	60.5	12.9	16.6	10.0	236	230	22.3	1.1	72.8	2.8	Diabetic at 19; retinitis, hypertension; peripheral arteriosclerosis with loss of toe
Hick.....	34	F	6.9	6.7	67.8	6.3	18.8	7.1	235	226	24.5	1.0	73.0	1.5	Diabetic at 19; mucous colitis; no vascular complications
Grim.....	29	M	6.9	6.9	60.6	11.8	20.2	7.4	276	254	25.2	3.8	70.0	1.1	Diabetic at 27; Kimmelstiel-Wilson Disease; pulmonary tuberculosis
Pool.....	55	M	6.6	6.7	57.9	14.5	17.9	9.8	299	268	25.2	1.9	71.7	1.2	Diabetic at 45; retinitis proliferans; peripheral arteriosclerosis; albuminuria
Reil.....	23	M	7.0	7.6	70.1	8.8	15.4	5.9	229	203	25.6	5.5	66.7	2.3	Diabetic at 16; undernourished; no vascular complications
Fay.....	70	F	6.8	6.9	59.3	14.3	16.7	9.7	226	204	26.1	1.8	69.1	2.9	Diabetic at 56; hypertension; diabetic gangrene
Weba.....	17	F	7.6	7.9	63.6	16.7	13.6	6.2	175	153	27.8	2.4	67.5	2.3	Diabetic at 16; no vascular complications
Manc.....	16	M	6.8	6.6	73.1	8.5	14.7	3.7	120	95	33.2	1.0	65.9	0.0	Diabetic at 14; moderately severe; no vascular complications
Maca.....	44	M	5.8	6.2	58.5	13.6	17.3	10.6	143	135	34.8	0.5	60.8	4.0	Mildly diabetic for 6 mo.; no vascular complications
Sent.....	50	M	6.1	6.3	64.0	9.7	14.8	11.4	267	252	34.9	0.1	62.6	2.3	Diabetic at 36; insulin sensitive; no vascular complications
Mare.....	21	M	5.5	6.2	62.5	10.7	18.6	8.3	276	252	37.7	4.1	56.8	1.3	Diabetic at 12; unexplained peripheral edema; no vascular complications
Nephrosis															
Dund.....	8	M	4.2	4.0	13.7	5.4	65.9	14.9	786	766	1.8	0.4	97.3	0.6	Duration 18 mo.; moderate edema; impaired renal function
Berl.....	4	F	4.1	4.3	16.5	1.3	82.2		595	543	2.6	0.0	97.4		Duration 9 mo.; moderate edema; normal renal function
Cour.....	28	M	4.1	4.8	36.0	3.5	33.8	26.7	731	678	3.3	0.0	93.9	2.7	Duration 1 mo.; massive edema; impaired renal function
Whal.....	1	F	4.9	4.6	32.5	10.4	34.8	22.4	564	531	3.8	7.0	87.5	1.8	Duration 3 mo.; massive edema; autopsy–glomerular nephritis
Lope.....	3	F	3.7	3.9	23.7	6.8	62.0	7.4	1150	1183	4.1	0.0	95.4	0.5	Duration 7 mo.; moderate edema; impaired renal function
Malo.....	30	M	4.8	5.0	43.7	5.5	34.2	16.6	580	519	5.1	0.0	93.5	1.4	Duration 5 mo.; slight edema; impaired renal function
Godn.....	2	F	5.3	4.4	37.0	5.2	44.5	13.4	556	531	5.3	0.0	92.6	2.4	Duration 8 mo.; slight edema; impaired renal function
Smit.....	55	M	4.8	4.8	45.5	5.7	33.1	15.4	659	571	5.4	0.5	92.0	2.2	Duration 3 mo.; massive edema; impaired renal function
Glaz.....	23	M	4.0	4.4	36.2	6.3	35.4	22.0	456	430	6.2	0.6	90.4	2.8	Duration 8 yr.; moderate edema; normal renal function
Sant.....	2	M	3.7	3.5	25.6	5.1	60.1	9.2	413	409	6.3	0.0	93.2	0.9	Duration 8 mo.; massive edema, normal renal function
Cain.....	16	F	3.7	4.1	48.4	4.3	41.1	6.1	439	406	8.1	0.9	90.8	0.3	Duration 18 mo.; moderate edema; impaired renal function
Para.....	14	F	4.9	4.4	45.3	10.1	30.7	14.0	390	354	8.7	1.0	88.0	2.3	Duration 9 mo.; slight edema; normal renal function

greater than in Lever's[3] or our own normals.[1] The amount of the alpha$_1$ globulin is, however, not great enough to change materially the direction of results or the inferences that may be derived from them.

There is indication that the separation of beta$_2$ globulin and fibrinogen between Fractions C and D is variable and incomplete both in normal and in pathologic plasmas. For this reason in subsequent discussion of protein fractionation these two fractions are considered together.

TABLE III

SUMMARY OF DATA ON DISTRIBUTION OF PROTEINS AND CHOLESTEROL

		Normal Young Men and Women Age 18–35	Normal Older Men and Women Age 45–65	Athero-sclerotics	Diabetics	Nephrotics
Total protein						
Sum of fractions (gm. %)	Average	7.3	7.3	7.3	7.1	4.4
	range	6.5–8.5	6.5–8.5	6.1–8.3	6.2–8.7	3.2–5.0
Protein in A						
Per cent of total	Average	69.9	65.7	60.6	62.0	33.7
	range	63.4–76.8	57.7–73.2	44.4–69.6	37.7–73.1	13.7–48.4
Gm. %	Average	5.12	4.79	4.42	4.38	1.49
	range	4.63–5.68	4.19–5.49	3.17–5.36	2.65–5.55	0.55–2.20
Protein in B						
Per cent of total	Average	11.6	13.1	13.3	11.7	5.8
	range	8.0–15.3	7.7–18.9	9.1–21.8	5.3–18.8	1.3–10.4
Gm. %	Average	0.86	0.96	0.97	0.83	0.25
	range	0.52–1.31	0.67–1.60	0.52–1.80	0.35–1.55	0.06–0.48
Protein in C						
Per cent of total	Average	11.6	12.8	14.7	17.3	43.2
	range	9.1–15.4	9.5–17.8	10.1–24.1	9.3–38.7	30.7–65.9
Gm. %	Average	0.86	0.93	1.08	1.22	1.83
	range	0.65–1.22	0.62–1.28	0.77–1.71	0.66–2.72	1.35–2.67
Protein in D						
Per cent of total	Average	6.9	8.4	11.5	8.6	15.3
	range	4.2–12.2	4.7–12.8	6.5–19.2	3.7–13.7	6.1–26.7
Gm. %	Average	0.50	0.62	0.85	0.61	0.68
	range	0.29–0.91	0.36–1.02	0.47–1.59	0.24–1.06	0.23–1.28
Total cholesterol						
Sum of fractions (mg. %)	Average	190	245	259	254	577
	range	125–258	163–340	135–452	95–794	354–1183
Cholesterol in A						
Per cent of total	Average	29.3	23.1	13.6	19.1	5.1
	range	13.3–46.1	11.7–40.7	5.6–25.6	5.6–37.7	1.8–8.7
Mg. %	Average	54.3	54.0	33.5	42.9	26.7
	range	32.4–96.1	31.8–80.0	9.3–55.3	14.3–94.9	13.9–48.1
Cholesterol in B						
Per cent of total	Average	3.1	1.6	2.2	1.6	0.9
	range	0–8.4	0–8.6	0–8.7	0–8.0	0–7.0
Mg. %	Average	6.3	3.9	5.7	3.6	4.4
	range	0–21.6	0–16.8	0–23.6	0–15.0	0–37.2
Cholesterol in C						
Per cent of total	Average	64.5	71.4	78.8	76.3	92.2
	range	44.9–84.5	54.2–83.3	61.0–86.4	56.8–91.4	87.5–97.3
Mg. %	Average	123.0	177.0	205.0	200.0	539.0
	range	69.6–201.9	86.0–273.0	86.0–383.0	63.0–316.0	311.0–1129.0
Cholesterol in D						
Per cent of total	Average	3.0	3.8	5.1	2.4	1.6
	range	1.0–11.3	1.0–12.2	0.9–16.8	0–7.2	0.3–2.8
Mg. %	Average	5.5	9.5	12.6	6.6	8.6
	range	1.4–22.7	2.3–30.2	2.0–42.6	0–24.7	1.6–18.6

RESULTS

Distribution of Proteins and Cholesterol. The determinations on the fractionation of protein and cholesterol of all groups are brought together in Table II which includes age, sex and a short note on the clinical condition of each of the individuals selected for study. The total protein as determined by the microKjeldahl procedure may be compared with the sum of the biuret values in the fractions. Percentages of protein in each fraction are expressed in terms of the sum of the biuret values. The cholesterol as determined in the plasma may be compared with the sum of cholesterol values of the fractions.

In Table III averages and ranges of protein and cholesterol for the various groups have been assembled for comparison with normal values. Total protein and cholesterol in this table represent the sum of the fractions. It will be seen that in those patients with atherosclerosis and diabetes the levels of total protein and of the proteins in Fraction B (gamma globulins) deviate little from the normal range. As would be expected from previously published work,[4] both are extremely low in the nephrotics.

In addition to these characteristic changes in nephrosis four other deviations from normal are apparent to a variable degree in the three groups, atherosclerotics, diabetics and nephrotics: (1) *Protein in Fraction A tends to be diminished.* (2) *Protein in Fraction C plus D tends to be increased.* (3) *Values for the total cholesterol average higher than normal.* (4) *The relative and absolute amounts of cholesterol in Fraction A tend to be lower with correspondingly higher values of cholesterol in Fraction C.*

Since there were only two patients with xanthoma tendinosum, they have been omitted from Table II and from later discussion. It will be noted in Table I, however, that they display the defect in lipid distribution to an extreme degree but that the distribution of proteins appears to be entirely within normal limits.

Averages and ranges are less informing than the actual distribution of the individual determinations.

Reduction of Protein in Fraction A: In Figure 1 the amount of protein in Fraction A is plotted against the amount of total protein for each individual analysis. Isobars of percentage are included for reference. Comparison is made between normal individuals, atherosclerotic patients and diabetics. Determinations in nephrotics are omitted from the diagram because the abnormalities both of total protein and of protein in Fraction A are so great as to make inconvenient their charting with values in other conditions.

It will be seen that, in general, the amount of

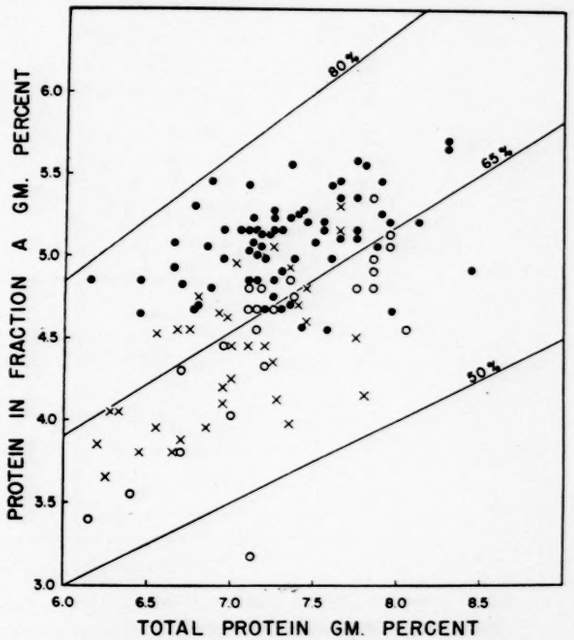

FIG. 1. Relation of protein in Fraction A to total protein (sum of biurets). ● Normal subjects; ○ atherosclerotics; × diabetics.

protein in Fraction A tends to rise in proportion to the total protein but varies widely at each total protein level. Among the normal individuals the variations are relatively small, the protein of Fraction A seldom constituting less than 65 per cent of the total and its amount never falling below 4.6 gm. per cent, even at the lower levels of total protein. In atherosclerotics and diabetics variations are much greater, with a few values not far above those in nephrosis and with others particularly among the diabetics that invade the normal range.

Increase in Protein in Fraction C + D: In Figure 2 the amounts of protein in Fractions C + D are plotted against the amounts of total protein for each individual determination. Again isobars of percentage are included for reference. As would be expected, values for the protein in Fractions C + D are roughly proportional to the amount of total protein. It is apparent, however, that there is great variability at each total protein level. Among the normal individuals the range

of values is within relatively narrow limits. In the younger group the highest values are 1.75 gm. per cent and 22.3 per cent of the total. Values higher than this are in the older men and women and have upper limits of 1.97 gm. per cent and 25.4 per cent of the total protein. and women. In this group the differences in the age groups of normal individuals as well as the differences between men and women may be seen.

The actual amounts of cholesterol in Fraction A are not shown in Figure 4. It is noteworthy,

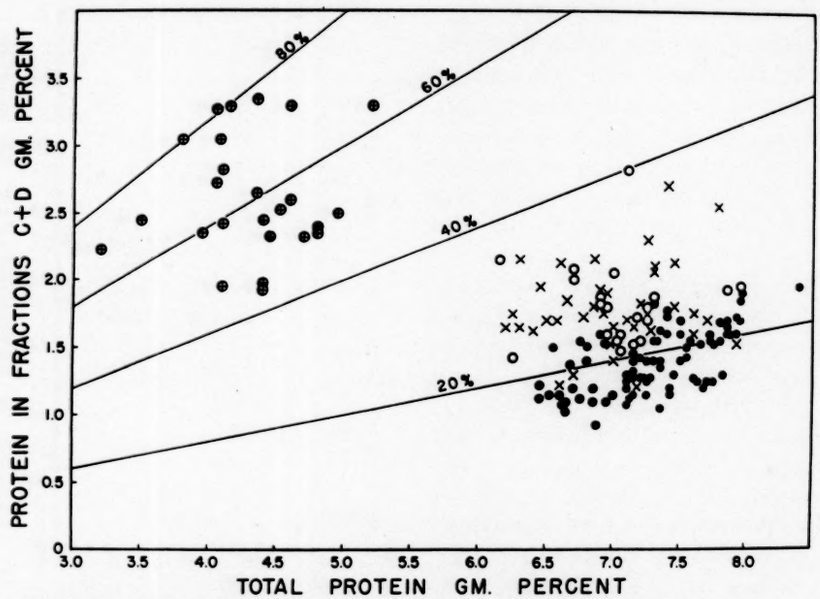

FIG. 2. Relation of protein in Fractions C + D to total protein (sum of biurets).
● Normal subjects; ○ atherosclerotics; × diabetics; ⊕ nephrotics.

Among the atherosclerotics and diabetics the variability of values is very great but with relatively few determinations that fall within the normal range.

It is to be especially noted that although in the nephrotics the total protein is extremely low the actual weight of the proteins of Fraction C + D exceeds that of other groups. (Table II.)

Higher Average of Total Cholesterol in Plasma: Figure 3 is a scatter graph to show the levels of cholesterol in the plasma of each group. It emphasizes the well known fact that in atherosclerotics and diabetics the higher average of cholesterol is attributable chiefly to the occurrence of a relatively few cases of hypercholesterolemia, the great majority of values falling within the normal range. In the nephrotics, as would be expected, hypercholesterolemia of high degree is uniformly encountered.

Decrease in Percentage of Cholesterol in Fraction A: Figure 4 presents the distribution of values for the percentage of the total cholesterol found in Fraction A in the various groups. A solid line is drawn arbitrarily at 20 per cent to represent an approximate lower limit of normal young men

however, that the average in each of the pathologic conditions is less than normal. Even the nephrotics, with their extremely high values for total cholesterol, have an average of only 26.7 mg. per cent as compared to the average for young normal men and women of 54.3 mg. per cent. The degree of diminution in the various groups may be seen by reference to Table II.

Statistical Significance: The statistical significance of the various changes in protein-lipid relationships in atherosclerosis and diabetes is indicated in Table IV. Nephrotics have been omitted because the values in all categories are outside of the range of the other groups. The table includes three smaller groups of patients for comparison with normal individuals of like age and sex. The number of cases in other categories was too small to permit statistical analysis.

Repeated Tests on Individuals: Some of the subjects were studied more than once over considerable periods of time. Usually abnormal patterns of protein and cholesterol distribution tended to persist even with considerable variations in diet, activity and life situations. An

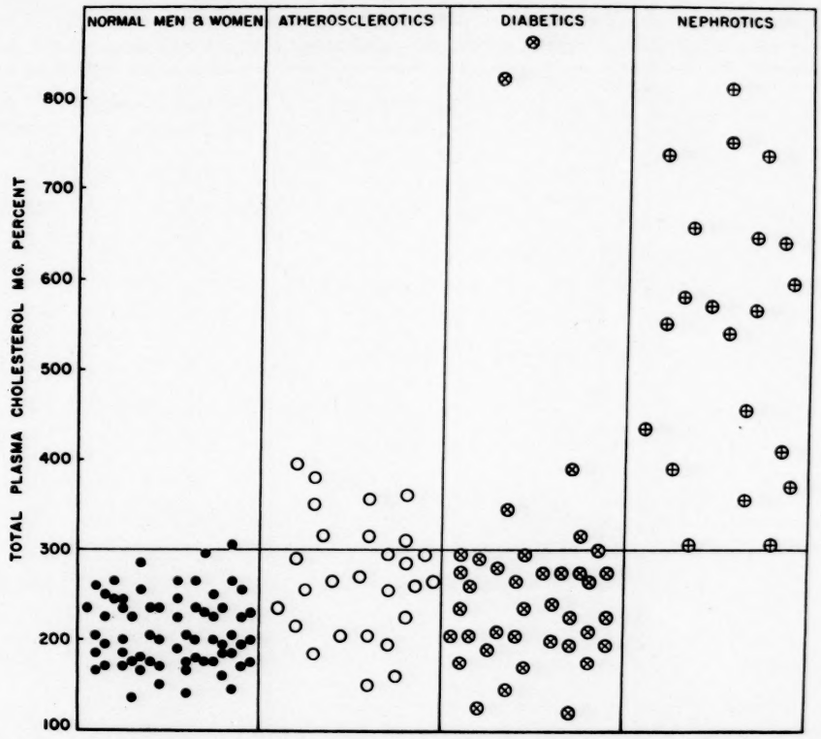

FIG. 3. Comparison of total plasma cholesterol in normal and pathologic states. ● Normal subjects; ○ atherosclerotics; ⊗ diabetics; ⊕ nephrotics.

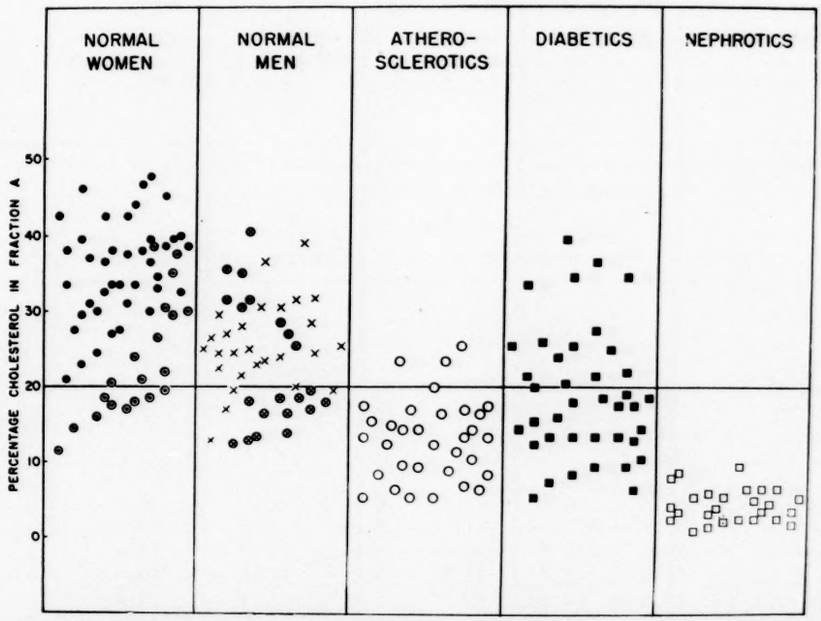

FIG. 4. Percentage of total cholesterol in Fraction A. ● Normal women, age eighteen to thirty-five; ⊙ normal women, age forty-five to sixty-five; × normal men, age eighteen to thirty-five; ⊗ normal men, age forty-five to sixty-five; ○ atherosclerotics; ■ diabetics; □ nephrotics.

TABLE IV

STATISTICAL ANALYSIS

Group	Sex	No. of Subjects	Per cent Cholesterol in Fraction A		Per cent Protein in Fraction A		Per cent Protein in Fraction C + D	
			Mean ± S.D.	P*	Mean ± S.D.	P	Mean ± S.D.	P
Normal subjects (age 18–65)	M + F	85	27.8 ± 7.20	$<10^{-9}$	67.8 ± 2.92	$<10^{-9}$	19.9 ± 1.88	$<10^{-9}$
Atherosclerotics (age 28–66)	M + F	33	13.6 ± 5.16		60.6 ± 6.10		26.1 ± 4.72	
Normal subjects (age 18–65)	M + F	85	27.8 ± 7.20	$<10^{-6}$	67.8 ± 2.92	$<10^{-6}$	19.9 ± 1.88	$<10^{-8}$
Diabetics (age 4–84)	M + F	34	19.1 ± 8.45		62.0 ± 6.58		26.3 ± 6.34	
Normal subjects (age 18–35)	F	20	34.3 + 6.35	$<10^{-9}$	69.1 ± 2.92	$<10^{-2}$	19.0 ± 2.09	$<10^{-2}$
Diabetics (age 16–36)	F	10	18.1 ± 6.75		60.3 ± 9.29		27.4 ± 9.72	
Normal subjects (age 18–35)	M	24	25.2 ± 6.48	$<10^{-9}$	70.5 ± 3.20	$<10^{-5}$	18.2 ± 1.75	$<10^{-5}$
Atherosclerotics (age 28–38)	M	10	12.4 ± 3.58		64.2 ± 4.30		24.0 ± 3.66	
Normal subjects (age 45–65)	M	21	22.9 ± 8.69	$<10^{-5}$	67.5 ± 3.32	$<10^{-6}$	20.0 ± 2.11	$<10^{-7}$
Atherosclerotics (age 45–65)	M	15	12.0 ± 5.43		58.3 ± 6.46		27.8 ± 5.37	

* P is the probability of chance difference between the means and is obtained for each difference from Tables I and II of FISHER, R. A. Statistical methods for research workers.

TABLE V

REPEATED FRACTIONATION IN A DIABETIC SUBJECT (MART)

Date	Total Protein (Gm. %)	Protein in Fractions (%)				Total Cholesterol (Mg. %)	Cholesterol in Fractions (%)			
		A	B	C	D		A	B	C	D
7/25/50	6.6	57.2	10.7	23.8	8.1	360	9.6	0.9	87.7	1.8
7/31/50	7.1	57.6	12.2	21.4	8.6	354	12.7	2.4	83.5	1.6
8/15/50	6.9	59.9	11.5	20.8	7.8	342	13.6	1.9	83.1	1.3
8/25/50	7.0	61.3	12.4	18.3	8.0	315	12.9	1.0	84.7	1.5
9/14/50	7.2	60.5	13.3	19.7	6.4	311	13.8	1.2	83.1	1.6
10/24/50	6.9	61.8	12.1	18.8	7.2	328	14.2	1.3	82.9	1.7
11/29/50	6.7	63.4	10.8	18.9	7.0	324	14.0	2.3	81.0	2.6
12/7/50	9.3	64.1	13.5	17.0	5.4	270	16.3	2.1	79.1	2.3
12/12/50	6.4	63.2	11.8	18.6	6.4	288	14.0	2.1	79.5	4.3
3/16/51	7.2	64.0	12.2	16.4	7.4	322	18.4	1.8	76.6	3.3

TABLE VI

PHOSPHOLIPIDS AND CHOLESTEROL-PHOSPHOLIPID RATIOS IN PLASMA AND FRACTIONS

Subject	Age	Sex	Total Plasma		Fraction A		Fraction C	
			mg. %	Chol./P'lipid.	mg. %	Chol./P'lipid.	mg. %	Chol./P'lipid.
Average of Normal								
			253	0.95	107	0.51	116	1.35
Atherosclerosis								
Harr	39	M	244	1.04	62	.34	135	1.29
Minn	27	M	225	1.01	63	.37	130	1.25
Loga	37	M	234	1.14	67	.49	150	1.38
Newc	32	M	333	1.04	76	.49	218	1.44
Duff	64	F	276	.96	79	.47	172	1.25
Sulp	53	F	287	.94	98	.53	143	1.41
Newm	45	M	282	1.13	57	.44	187	1.41
Stow	66	M	267	.95	58	.31	185	1.21
Sals	38	M	328	1.01	80	.38	206	1.35
Bowl	48	M	251	1.03	68	.54	141	1.32
Schu	60	M	297	.81	101	.42	147	1.13
Stoc	53	M	238	1.07	68	.47	137	1.41
Sapr	57	F	300	1.03	87	.47	170	1.38
Mole	37	M	261	1.12	100	.52	131	1.47
Mean			273	1.02	76	.45	161	1.34
Diabetes								
Webb	64	F	294	.91	84	.41	165	1.26
Mart	21	F	310	1.18	94	.49	179	1.52
Smit	42	M	661	1.24	86	.68	491	1.26
Gust	24	F	602	1.43	70	.64	451	1.61
Faja	59	M	230	.81	79	.40	135	1.06
Sull	45	M	214	.91	110	.51	75	1.50
Mean			385	1.08	87	.52	249	1.37
Nephrotic Syndrome								
Glaz	23	M	377	1.21	48	.56	272	1.43
Cour	28	M	552	1.33	52	.44	424	1.50
Smit	57	M	522	1.44	46	.45	433	1.50
Saut	2	M	356	1.19	58	.45	268	1.43
Malo	30	M	461	1.26	61	.43	353	1.37
Berl	4	F	446	1.33	27	.51	375	1.41
Lope	3	F	736	1.17	17	.54	647	1.29
Mean			493	1.28	44	.48	396	1.42
Biliary Obstruction								
Peys	68	F	1595	.49	1225	.44	268	.53
Meek	54	F	765	.57	379	.42	343	.61
Kurt		F	708	.58	338	.50	213	.80
Mean			1023	.55	647	.45	275	.65

example is offered in Table v, which includes tests performed over the course of eight months on a twenty-one year old girl who had had diabetes since the age of ten. She had never been under satisfactory therapeutic control and had suffered many complications including episodes of severe ketosis, insulin reactions and severe local infections. At the time of the first examination she appeared to be in an excellent state of nutrition and exhibited no ketosis or polyuria. She had, however, a few small retinal hemorrhages and an occasional faint trace of albumin was detectable in her urine. Following the first fractionation of her plasma with its disclosure of hypercholesterolemia and abnormalities in the distribution of protein and lipid she was hospitalized and placed on a strict diet with increase in the intake of protein, diminution in the intake of fat and a larger insulin dosage. This regimen, with the closest possible attention to her management, was continued for a period of three months. Later tests taken over a period of five months represented values when she was out of the hospital under severe emotional stress and only partial diabetic control. Over the entire period a tendency to correction of the abnormal pattern is apparent but the essential defects in distribution are persistent.

Distribution of Phospholipid and the Cholesterol-Phospholipid Ratios. In Table VI data concerning phospholipids and cholesterol-phospholipid ratios in pathologic states have been assembled for comparison with similar determinations in normal individuals.

In patients with atherosclerosis and diabetes the average phospholipid is slightly above that of the normal group. As with cholesterol this is due to the presence in the series of a few patients with markedly elevated levels. The distribution of phospholipid roughly parallels that of cholesterol; the percentage is decreased in Fraction A and increased in Fraction C. In the patients with nephrotic syndrome the mean plasma phospholipid is double that of the normal and the distribution deviates greatly with a marked reduction of phospholipid in Fraction A and a more than threefold increase in Fraction C.

The plasma cholesterol-phospholipid ratios are not significantly different from the normal in any group except the nephrotics, in whom a striking increase is apparent. In atherosclerotics, diabetics, nephrotics and in the one patient with xanthoma tendinosum who was tested the cholesterol-phospholipid ratios of Fraction A and Fraction C are similar to those found in normal subjects. The values of some individual cases rose above the normal levels but the averages for each group fall closely within the normal range. Indeed, in a considerable range of miscellaneous pathologic conditions in which these values have been determined no significant variation was found except in patients suffering from biliary obstruction. In this disease the ratio for Fraction A is similar to that of all other states but the ratio in Fraction C is very low and in one case is only a little higher than the ratio for Fraction A. In Table VI values for three patients with biliary obstruction have been included for comparison. They are of special interest in this study since they represent instances of hypercholesterolemia in which a tendency to atherosclerosis is thought to be absent.

COMMENTS

The pattern of deviation from the normal is similar in all groups and is evident both in those who have suffered the consequences of atherosclerosis and those who were thought because of their various diseases to be susceptible to its formation. Grossly, the nephrotics differed from the others only in degree of abnormality and in their tendency to extreme reduction of total protein and gamma globulins. In considering the various deviations we are inclined to regard the diminution in cholesterol content of Fraction A as the most significant since it implies a reduction in alpha lipoproteins and a relative increase in beta lipoproteins. It is evident, however, that this deviation is seldom marked without an accompanying reduction in the protein of Fraction A as well as significant and often large augmentation in the proteins of Fractions C + D. Although the various changes do not always parallel each other and could be regarded as independent variables, it is not possible to discuss one deviation without some cognizance of the others.

Influence of Age and Sex. Our studies of normal subjects indicated that exact comparison of individual patients can be made only with normal persons of the same age and sex. Although the number of cases in this series does not permit exact matching, some comparison of groups were made in Table IV. Among the atherosclerotics there were ten men between the ages of twenty-eight and thirty-eight, and fifteen men between the ages of forty-five and sixty-six. Among the

diabetics ten women were studied between the ages of sixteen and thirty-six. Although the number of cases in each group is small, the deviations are so large as to leave no doubt of the significance of the abnormalities or of their lack of dependence upon age or sex.

Degree of Uniformity in Abnormalities in Protein and Lipid Relationships. Every case of the nephrotic group exhibits reduction in albumin, increase in the proteins in Fraction C + D and abnormalities in the distribution of lipids between the two groups of lipoproteins. All of the changes are of extreme degree.

In the atherosclerotic group abnormalities of some degree or kind were absent in only two cases. Marr, a woman of forty-two who had had an anterior wall infarction two years before her study, displayed no significant deviation from normal. Ober, a man of forty-one who had had a myocardial infarction a year before the observation, also presented a fractionation entirely within normal limits. Two other patients, Mysz and Bruc, who had more than 20 per cent of their cholesterol in the form of alpha lipoprotein, displayed reduction in albumin and increase in the protein of the C + D fractions.

The results in the diabetics were less consistent. Of the total group only a few, however, were completely free from abnormal tendencies. Although eleven of the thirty-four cases displayed percentages of cholesterol which might have been regarded as essentially normal for their age and sex, only four failed to show abnormal tendencies in protein distribution. The first and most normal of this small group was Graf, a man of forty, with very mild diabetes usually requiring no insulin. Reil, a man of twenty-three, had had diabetes since the age of sixteen. He displayed no stigmas of vascular damage. Marc, a boy of sixteen, with moderately severe diabetes for two years and with no vascular complications, was distinguished only by an extremely low cholesterol of 120 mg. per cent. Weba, a girl of seventeen, had shown high insulin requirement but presented no evidence of vascular complications.

Other cases of diabetes are worthy of note because their deviations from normal were much less than might have been expected from their clinical condition. Pool, a man of fifty-five, had a percentage of cholesterol in Fraction A essentially within normal limits. His abnormality consisted only in a reduction in albumin and a marked increase in the protein in Fraction C + D. Clinically, however, he had retinitis proliferans with partial blindness, albuminuria and hypertension. Fay, a woman of seventy who had had an amputation for diabetic gangrene, presented other evidences of peripheral atherosclerosis but displayed a percentage of cholesterol in Fraction A within normal limits, with only moderate reduction in albumin and moderate increase in the protein in Fraction C + D. Wrab, a man of forty-one who had had diabetes for twenty-one years, showed retinitis, hypertension and peripheral arteriosclerosis which had caused the loss of a toe but exhibited only moderate deviations from normal in the distribution of his proteins.

In reviewing the group of diabetics, therefore, discrepancies are apparent in the findings in a number of individuals, some who were quite young displaying extreme abnormalities while others showed deviation which cannot be regarded as very significant. It was hoped at first that it might be possible to differentiate diabetics who have a tendency to atherosclerosis from those who do not. Thus far the data do not support this hope. It is apparent, however, that from the criteria of this investigation the diabetic group cannot be regarded as statistically homogeneous.

Significance of Abnormalities in Protein Distribution. The reduction of the protein in Fraction A must be attributed chiefly to a diminution in the amount of albumin. This is evident from our electrophoretic analyses and in diabetes has been demonstrated by Schneider, Lewis and McCullagh.[5] Contributory, also, is the diminution in the amount of alpha lipoproteins. Our present knowledge of the composition of these compounds is insufficient to permit determination of their exact concentration in Fraction A. By the most liberal calculation, however, they can account for only a small fraction of the total in many of the cases.

The increase of protein in Fractions C + D is due in part to a greater concentration of beta lipoproteins. The amount of these substances may be approximately calculated from Oncley's analyses which indicate that, on the average, cholesterol constitutes 31 per cent of the beta lipoprotein molecule.[6] By use in the calculation of the cholesterol values actually determined in Fractions C + D it can be readily shown that the increased amount of protein is attributable only in part to beta lipoproteins. Electrophoretic analyses indicate (Table III), also, large incre-

ments in alpha$_2$ globulin and fibrinogen. It is possible but not easily demonstrable that beta$_1$ lipid-poor globulins also constitute a considerable part of the total increment.

Significance of Abnormalities in Lipid Distribution and Their Relation to the Pathogenesis of Atherosclerosis. For many years the idea has been entertained that disturbances of cholesterol and other lipids in the plasma might play an important role in the pathogenesis of atheroma. For some time it was believed that the level of plasma cholesterol might be indicative of a tendency to the disease, particularly since it was often elevated both in clinical and experimental atherosclerosis. Recently there has been a revival of interest in the observation of Boyd[7] that phospholipids play a role in the stabilization of the plasma lipid emulsion. Ahrens and Kunkel,[8] and Kellner, Correll and Ladd[9] have by different approaches emphasized the possible significance of the cholesterol-phospholipid ratio. Gertler, Garn and Lerman[10] in a careful statistical study compared the plasma of normal young men with that of patients of like age who have survived coronary sclerosis. They found averages of cholesterol-phospholipid ratios significantly higher in the latter but encountered many individual cases in which the ratios were within normal range. Gofman and his associates,[11] by study of the flotation of lipoproteins in the ultracentrifuge, have shown that atherosclerotics have a high concentration of certain lipoprotein molecules (S_f 10 − 20) more frequently than normal individuals. The nature of these molecules is not exactly known but they may form a small fraction of the large group of beta lipoproteins.

In this study we have shown, as have many others, that the average cholesterol level in the plasma of atherosclerotics is higher than normal but, also, that the high average is caused by inclusion in the group of a relatively small number of patients with marked hypercholesterolemia. Most atherosclerotics exhibit plasma cholesterol values well within the normal range. We have shown disturbances in cholesterol–phospholipid ratios. Our analyses indicate clearly that a high percentage of alpha lipoprotein, such as is found in normal young women, will tend to produce a relatively low cholesterol–phospholipid ratio in the plasma and that the low values of alpha lipoprotein in atheroma and kindred conditions will tend to increase the ratios. On the other hand, the highly variable ratios in both Fraction A and Fraction C distort this simple relationship. It is apparent that cholesterol–phospholipid ratios in unfractionated plasma cannot be predicted by determination of the distribution of cholesterol between the two types of lipoproteins. The cholesterol–phospholipid ratios of the plasma were, like those of Gertler and Garn, somewhat higher than normal in the atherosclerotics. The variation in individual patients, however, was great and it seems unlikely that this factor alone can have a significant pathogenetic influence in the disease.

The outstanding fact in our own observations is the relative and absolute reduction in alpha lipoprotein in atherosclerosis and kindred conditions. This is particularly apparent in the hypercholesterolemic states of nephrosis and xanthoma tendinosum in which, in spite of enormous increases in total cholesterol, the concentration of alpha lipoprotein may be much less than in normal individuals. Under such circumstances there is always a relative and absolute increase in the concentration of beta lipoproteins. It is suspected, although as yet not proven, that the S_f 10 − 20 molecules of Gofman form a part of the beta lipoprotein increment.

In speculating upon possible mechanisms it has occurred to us that the presence in higher than normal concentration of beta lipoproteins may in itself be significant in the genesis of atheroma. Furthermore, it might be suspected that all of the group rather than the small fraction of Gofman's S_f 10 − 20 contributes to a tendency to instability of lipid emulsions in plasma and tissue fluids.

Relation of Changes in Protein and Lipid Distribution to the Diagnosis of Atherosclerosis. At this stage it would be premature to suggest that the changes in protein and lipid distribution could be used in the early recognition of atherosclerosis. Thus far we have not explored sufficiently the pattern in other pathologic conditions to claim specificity of the changes for an atherosclerotic tendency or to exclude the possibility that they may represent a general response of the body to a variety of diseases or insults. The influence of such factors as infection, malnutrition, dietary excesses, gonadal hormones and other endocrine influences have not been fully explored. Furthermore, in this small series we have encountered several cases in which the clinical evidences of atherosclerotic complications were unequivocal but which did not exhibit characteristic abnormalities in the plasma.

Nevertheless the data are most suggestive of

a more than coincidental association. Thus far we have not seen any indication of the defect in healthy young women. We have encountered reduction in the alpha lipoprotein and relative increase in beta lipoprotein in only a few vigorous, apparently healthy young men. We have seen extreme changes in young diabetics of both sexes before any clinical evidence of vascular complications could be detected. It may be emphasized, also, that the association of changes in protein and lipid distribution in survivors of myocardial infarction or those who otherwise show the consequences of advanced atheroma is more constant than the presence of hypercholesterolemia or the significant elevation of cholesterol–phospholipid ratio in the unfractionated plasma.

This phase of the subject merits extensive further study. The results of this communication indicate clearly that future attempts to relate lipid of the plasma to the deposition of lipid in tissue must take account of their combinations with protein.

SUMMARY AND CONCLUSIONS

1. Patients who have survived coronary occlusion or present otherwise unequivocal evidence of the complications of atherosclerosis frequently exhibit several abnormalities in the distribution of proteins and lipids in the plasma. These include a tendency to reduction of albumin and alpha lipoprotein and a relative and absolute increase in beta lipoprotein, as well as other components of Cohn's Fractions I and III.

2. These changes may be apparent without hypercholesterolemia or recognizably significant elevation of the cholesterol–phospholipid ratio of the unfractionated plasma. Like normal individuals, patients in the atherosclerotic group exhibit, in the fraction containing alpha lipoproteins, cholesterol–phospholipid ratios which average around 0.50 and in the fraction containing beta lipoproteins ratios which range about 1.40.

3. Similar changes in protein and cholesterol distribution are apparent in conditions which are known to predispose to early and extensive atherosclerosis. In many diabetics they are apparent before any vascular complications of the disease are clinically recognizable. They were seen in two cases of familial xanthoma tendinosum. They are present to an extreme degree in patients with the nephrotic syndrome.

4. It is suggested that future attempts to relate lipids of the plasma to the deposition of lipids in tissue must take account of their combinations with protein; also, that further exploration of protein-lipid relationships may be rewarding both in clarifying the pathogenesis of atherosclerosis and in aiding in its early recognition.

Acknowledgments: We gratefully acknowledge the analytical determinations performed by Miss Hanna Fisher and Miss Mary Donahue.

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Combined Staff Clinic

Hypersplenism

THESE are stenotyped reports of combined staff clinics of the College of Physicians and Surgeons, Columbia University, and the Presbyterian Hospital, New York. The clinics, designed to integrate basic mechanisms of disease with problems of diagnosis and treatment, are conducted under the auspices of the Department of Medicine. The reports are edited by Dr. Gilbert H. Mudge.

DR. JOSEPH C. TURNER: This clinic will consider four aspects of hypersplenism: (1) the concept of hypersplenism, (2) the role of the splenic circulation, (3) the clinical aspects of thrombopenic purpura, with a consideration of splenectomy and cortisone therapy, and (4) a consideration of acquired hemolytic jaundice and splenic neutropenia and their management. Our plan is first to examine the theory of hypersplenism and then to pass on to an examination of certain clinical and experimental data which bear upon the general problem of splenic function.

It was Eppinger who some forty years ago offered an analysis of hemolytic anemias in terms of increased blood destruction and phagocytosis within the spleen. A few years later Kaznelson applied analogous ideas to the problem of thrombopenic purpura; splenectomy was then successfully undertaken in this disease. More recently Doan and others have extended the concept of hypersplenism to include so-called splenic neutropenia emphasizing that, according to their view, the mechanism common to all of these conditions involves sequestration of the formed elements of the blood in the splenic pulp with subsequent phagocytosis by hyperactive reticuloendothelial cells. It is a corollary of this idea that the bone marrow is normally active or even hyperplastic. There is now a considerable body of morphologic as well as other kinds of evidence to indicate that the spleen may indeed selectively retain abnormal red cells and accelerate their destruction. The pathway of destruction is not, however, a matter on which there is general agreement. Furthermore, it may be that the spleen exercises other effects on the hematopoietic tissues and blood cells. Before dealing with these questions it will be best to recall briefly the well established functions of the spleen in health.

You are all aware of the hematopoietic function of the spleen in lower vertebrates such as fishes and Amphibia. In frogs, for example, the spleen is the principal site of blood formation most of the year. After hibernation or after splenectomy the marrow may become red. In mammals the spleen is active in forming red cells during part of embryonic life but in the normal adult it may be viewed as a kind of phylogenetic remnant—a highly vascularized mass of lymphocytes and reticuloendothelial cells (macrophages) with a peculiar circulation. We do not propose to consider today the pathologic conditions under which the spleen resumes the formation of red cells and myeloid cells but remind you of this function largely to point out that the spleen possesses whatever such potentialities there may be in the lymphocyte and macrophage (reticuloendothelial) systems.

A second function of the spleen is that of blood storage. In some lower animals an appreciable and physiologically significant amount of blood may be deposited in the spleen to be called out into the circulation under stress. The organ in normal man is so small that it can hardly contribute materially to the restoration of blood volume, as after hemorrhage for example. In a sense, though, it is this depot function that is taken to be perverted in hypersplenism.

Beyond this, evidence for special activities of the normal spleen is slim. It may be that destruction of blood cells takes place but it seems probable that this amounts to no more than phagocytosis of fragments resulting from the process of intravascular hemolysis. Animal experiments indicate that splenectomy has little or no influence on the life span of the normal red cell.

Turning now to the spleen in human disease, we begin with the problem of segregation of blood cells. More than a decade ago Klemperer proposed an interpretation of the histologic findings in hemolytic jaundice that has been entirely borne out by subsequent clinical

studies. Dr. Sproul may say more of this, and for the moment it is enough to state that Klemperer's view was that in hemolytic anemia red cells were indeed trapped in the spleen and that this occurred because the cells were abnormal. Apart from the morphologic grounds in support of this view we now have several kinds of evidence bearing on this question.

In the first place it appears that when granules, e.g., starch, of different sizes are injected intravenously in an animal those above a certain critical diameter, about 5 μ, are retained in the spleen in great numbers while smaller particles pass through more or less freely. Following this lead Castle and others have been able to show that the relatively thick spherocytes of hemolytic jaundice may be segregated and withheld by the spleen. In anticipation of splenectomy a patient was transfused with cells from a donor of a different blood group. The relative numbers of donor's and recipient's cells in any mixture of the two could be determined by serologic methods. The spleen was removed. The blood of the spleen and that of the peripheral circulation were compared in terms of the ratio of patient's cells to donor's cells (normal cells). This ratio could be appreciably altered if the spleen retained one type of cell (spherocyte) preferentially. And indeed, so it proved. The ratio of patient's to donor's cells was higher in the spleen.

There can be little doubt that such segregation of abnormal red cells by the spleen is concerned in accelerated erythrocyte destruction. It is of some interest that the spleen need not be itself "hypersplenic" for this to happen. Thus if the erythrocytes of congenital hemolytic disease are transfused into an intact normal subject, they are found by Ashby's technic to have a short life span, two or three weeks as opposed to the some seventeen weeks of life enjoyed by a normal red cell. On the other hand, if the recipient, otherwise normal, has been splenectomized, the intrinsically defective cells infused into him will survive almost as long as normal cells do.

Thus far we may conclude that hypersplenism, insofar as it is exemplified in congenital hemolytic jaundice, almost certainly involves segregation of abnormal red cells by the spleen and that this is associated with accelerated blood destruction. Since the spleen of normal subjects treats the red cells in much the same way, it is doubtful whether the prefix "hyper" is altogether appropriate.

In acquired hemolytic jaundice it is now evident as a result of the work of Coombs and other British investigators that the erythrocyte is rendered abnormal through the operation of an autoantibody which coats the cell. Coombs' test for this immune reaction has become familiar in its application to the demonstration of antibodies in erythroblastosis; the kind of antibody concerned in acquired hemolytic jaundice appears to be, at least superficially, similar in its physicochemical characteristics but has nothing to do with blood groups. Coombs' test for the presence of antibody on the red cell surface appears to be positive in most cases of acquired hemolytic jaundice as opposed to congenital hemolytic disease.

In acquired hemolytic jaundice abnormal red cells doubtless also are sequestered and destroyed in the spleen. But in this case we know that the primary etiologic factor is an agglutinin which transforms the erythrocyte. Should this kind of disease mechanism be called hypersplenism? Dr. Ranney will have more to say about this aspect of the subject.

Now we must get on to a few curious observations which suggest that the spleen is not simply a phylogenetic remnant capable merely of separating abnormal cells but may exercise a unique function. A good deal has been written about the role of the spleen in resistance to infection. It seems probable that the macrophages of the spleen may be useful in taking up bacteria, and that they and the other cells in the spleen, e.g., lymphocytes, may contribute to antibody formation. These actions may account in large part for the claims that splenectomy in animals often results in increased susceptibility to infection. Of more interest, perhaps, are the latent infections that may be made manifest by removal of the spleen. These include, according to the review some years ago by Perla and Marmorsten, "The Spleen and Resistance," (1) bartonellosis in six or more species, (2) eperythrozoon disease of mice, (3) piroplasmosis in dogs, cattle, etc., (4) malaria in apes and monkeys, (5) coccidiosis of the kidney in mice and (6) a spirochetal disorder of squirrels. For all of these and perhaps more it may be said that an apparently healthy animal is converted into a morbid one by removal of the spleen.

Now, it is rather odd that the majority of these infections are intimately related to the erythro-

cyte. By this I mean that the organisms of bartonella, eperythrozoon disease, piroplasmosis and malaria are found for the most part or exclusively in or on the red cell. Take, for example, the case of malaria. Years ago it was shown that if apes were inoculated with a species of malarial parasite and allowed to recover from a succession of paroxysms they remained well until splenectomized. The operation was followed by a recurrence of malarial infection which now ran a chronic rather than an episodic course. Recent studies of a similar kind in monkeys show much the same thing. Although the experimental disease in intact animals consists of a more or less limited number of episodes like the ones seen in tertian malaria in man, removal of the spleen is followed by a most unorthodox course of events. Continuous and heavy parasitization of the red cells of the peripheral blood ensues which goes on without remission for many months. Furthermore, the virulence of the organism for intact, normal monkeys is modified. Can such findings be taken to indicate some special relation between the metabolism of the red cell and the spleen?

For the human being some such relation is perhaps suggested by the morphologic changes in red cells observed after splenectomy. Of greatest interest are the so-called siderocytes that may sometimes although irregularly appear in the peripheral blood within forty-eight hours after removal of the spleen for conditions so diverse as purpura, sickle cell disease, hemolytic jaundice and traumatic rupture of the spleen. These cells contain in or on them small bodies about the size of the organisms of bartonella, but differing from the infectious agents in many respects. The differentiating feature responsible for the name "siderocyte" is the demonstration of considerable amounts of iron in the erythrocytic inclusions.

There are strains of mice whose erythrocytes contain entirely similar bodies. These are seen in the embryo and in newborn mice only. In such a case could it be that there is a relation between the siderocyte and the familiar fact that fetal hemoglobin differs chemically from adult hemoglobin?

But such questions lead us away from the main topic, and we must return to the more or less hard facts of human disease. Dr. Sproul will present the current views of the splenic circulation and its aberrations in disease.

DR. EDITH E. SPROUL: As I understand my obligation this morning, it is to present those features of structure and circulation of the spleen which may contribute to our understanding of its function of blood destruction.

The gross framework of the spleen is formed by a fibromuscular capsule and intercommunicating trabeculae, capable of contraction with the expulsion of a large portion of the blood held within the organ. Although this is not of importance in restoration of blood volume in man, such a mechanism in the normal spleen prevents the blood cells from remaining over-long in the intricacies of the pulp, and loss of contractile power adds to the sluggishness of the blood as it traverses this organ.

The splenic artery enters at the hilum and branches through trabeculae until it reaches a diameter of about 0.2 mm. It then bears a collar of lymphoid tissue, the Malpighian follicles, which it loses when it is reduced to a diameter of 40 to 60 μ. It is then surrounded by pulp as it gradually becomes a delicate capillary with only a single layer of endothelium between the flowing blood and pulp cords. It is generally believed that these capillaries open directly into a sponge-like reticulum meshwork, the pulp cord, which forms the bulk of the splenic tissue. The blood infiltrates slowly into this intricate mesh; part of the plasma is removed by way of lymph channels, concentrating the blood cells as they remain in the pulp. The blood ultimately passes into venous sinusoids which have a curious and unique structure. Their walls are formed by elongated endothelial cells placed in parallel rows in the direction of the flow of blood, with long narrow stomas between, much as the staves of a barrel are arranged. The sponge-like reticulum is directly applied to the sinusoidal walls so that the cells of the blood are filtered through these slit-like openings as they pass from the pulp into the venous sinuses and on into the veins of the organ. When the spleen contracts, blood is forced from the pulp into the sinuses, the lacunas of the reticular pulp are reduced about one-third the size they are in the relaxed state, and the blood temporarily takes a more direct route from arterial capillary to venous sinus. (Fig. 1.)

It should be understood that this description represents merely the current, most widely accepted view based on several sources of information. The histologists have observed

structure compatible with some such mechanism but we are dependent upon more dynamic studies for our concept of the passage of blood through the organ. Because of the complicated structure and indirect pathway, injection experiments have not been as successful as in other organs and have yielded opposing data. Artefacts related to contraction or relaxation of the organ are difficult to avoid. Knisely offered a somewhat different story based on direct microscopic study of the intact living spleen by means of quartz rod illumination but this was modified by MacKenzie who was able to keep the spleen under prolonged uninterrupted observation with continuous anesthesia. His conclusions were essentially as presented here and are most widely supported today.

The cellular content should be touched on if we are to discuss the functions of the spleen. The lymphoid follicles are not unlike those elsewhere in the body except for their relationship to the arterial tree, and they reflect all the changes occurring generally in lymphoid tissue. The pulp includes neutrophils, plasma cells and eosinophils, whose numbers vary with infectious diseases, but the majority of the pulp cells are those belonging to the reticuloendothelial system. It should be emphasized then that the blood cells during their slow passage through the pulp remain in intimate and relatively long contact with reticulum cells. These include primitive mesenchymal tissue capable of differentiation into hematopoietic cells, phagocytes, fixed reticulum cells and fibroblasts as well as endothelium of the sinuses. If there is a special cell worthy of being called a splenocyte, I do not know what it is and see no reason for postulating its presence. The relationship of blood cells to reticulum cells in the spleen is a unique one and we need not seek a special type of cell to explain the disintegration and phagocytosis of red blood cells which may occur more strikingly here than elsewhere in the body.

Abnormal circulation in the spleen might be expected to be accompanied with alteration in the pattern of blood cell destruction, and this is true under certain circumstances which we shall examine. When there is acute passive congestion of the spleen, the lacunas of the pulp are enlarged and the stomas of the venous sinuses are wider. The free passage of cells then somewhat counterbalances the obstruction due to venous congestion. When, however, there is persistent and severe venous hypertension for a prolonged

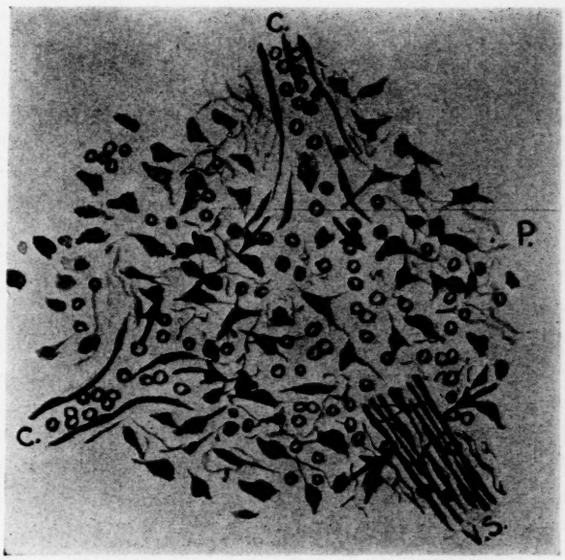

FIG. 1. Schematic drawing of most widely accepted concept of circulatory system in the normal spleen. C, arterial capillaries; P, pulp composed of meshwork of delicate reticulum fibers and cells of the reticuloendothelial system; V.S., venous sinus.

period, changes in structure occur in the spleen which alter the entire character of its contact with the blood. It becomes enlarged and very firm, due in good part to a pronounced increase in reticulum and collagen so that the sponge-like pulp is converted into more compact tissue now incapable of receiving the circulating cells because lacunas or intercellular spaces virtually cease to exist. Reticulum cells are also increased in number throughout the organ. The blood then passes more directly from arterial capillary through the solid pulp in channels which become lined by prominent endothelial cells throughout their length. The slit-shaped stomas of the venous sinusoids are now being boarded up by the collagenous tissue of the pulp, converting the entire splenic circulatory channel into a closed system as it is elsewhere in the body. (Fig. 2.) These new vascular channels become very numerous and prominent, containing a large quantity of blood which is no longer aided in its passage through the spleen by contraction of the organ since the capsule and trabeculae now have more collagen than muscle and the fibrosed pulp cannot be compressed. The lymphoid follicles undergo atrophy and fibrosis. These changes are often accompanied with hematocytopenia, which we sometimes refer to as Banti's syndrome. Any explanation of the more rapid disappearance of formed elements of the

FIG. 2. Schematic drawing of changes in circulatory system of spleen affected by portal hypertension. C, arterial capillaries now communicating directly with (V.S.) venous sinus; P, reticulum meshwork of pulp replaced by dense collagenous fibers with increased number of hypertrophied reticulum cells.

blood as they pass through such a spleen must be compatible with the observations as described. These can be summarized as greater stagnation of blood now in closed channels, removed from direct contact with reticulum cells. At the same time there is an increase in number of reticulum cells while the lymphoid tissue is markedly reduced. If then the reticulum cells exert any effect on the blood cells, it must be by humoral means rather than by increased phagocytic activity.

Under what circumstances does splenic vein hypertension lead to those changes clinically manifest as Banti's syndrome? Any form of complete obstruction of splenic or portal vein, especially by thrombi, benign tumors, scar tissue or even incomplete obstruction such as obtains when thrombi are recanalized in the form of cavernous transformation, is likely to do this. Intrahepatic portal vein obstruction such as results from cirrhosis provides a variable degree of hypertension depending in good part on the pattern of the lesion in the liver. Cirrhosis related to a deficiency state is apt to be generalized and uniform throughout the organ. Those liver cells receiving mainly portal venous blood are destroyed in larger numbers than those supplied chiefly by hepatic arterial blood. In consequence, scarring and profound obstruction of the portal system ensue. Cirrhosis following liver cell loss due to infectious or toxic agents is usually less uniform and less fibrotic, hence the degree of portal hypertension is commonly not adequate for Banti's syndrome to occur. Biliary cirrhosis is accompanied with little portal hypertension as a rule, but rarely the syndrome may develop. One of the most severe forms of intrahepatic portal obstruction is initiated by schistosomiasis and the splenic changes we have described with hematocytopenia regularly accompany the late stages of this infestation.

Hepatic vein thrombosis is usually not associated with this type of congestive splenomegaly. Right heart failure is usually of intermittent character and also exerts its greatest effect on the liver rather than the spleen.

Does Banti's syndrome (that is, the fibrosed spleen with closed circulatory system and hematocytopenia) occur alone without portal hypertension? I do not believe that it does. It is often very difficult indeed to find an obstruction which may occupy only a short segment of the vein; however, in all instances in which the entire system can be explored at necropsy the causative block can be found. Unfortunately it has been difficult to reproduce the entire picture in experimental animals because portal hypertension is not readily established. Introduction of silica into the portal radicles of the liver, however, may be followed by increased pressure in the portal vein and by splenic changes similar to those in man.

We have discussed this syndrome in some detail since it represents one of the simplest situations in which hematocytopenia may occur. In other conditions in which hematocytopenia of one type or another develops there is no uniform pattern in the spleen. Of these spherocytic anemia is most readily understood. The structure and circulatory pattern of the spleen remain unaltered but the meshwork of the pulp becomes crowded with spherocytes which are selectively filtered out by the slits in the sinusoidal walls. Presumably the biconcave normal red cell can pass through stomas of this contour more readily. Certainly the delayed passage of the spherocyte through the pulp facilitates its destruction and contributes to the anemia.

In thrombocytopenia purpura there is no consistent histologic abnormality. Doan supports the claim that there is increased phagocytosis of platelets in the spleen. Many careful observers are not able to corroborate this. Dameshek points to the bone marrow as the site of the trouble, describing the megakaryocytes as im-

mature with inadequate discharge of platelets. Improvement after splenectomy, he postulates, is due to removal of a humoral factor elaborated by the spleen which affects the bone marrow. This has never been satisfactorily demonstrated. Von Haam does not find any more phagocytosis of platelets in spleens removed surgically or at necropsy from patients with idiopathic thrombocytopenic purpura but does consider the number of reticulum cells present in such spleens to be more numerous, sometimes forming compact nodules. This, also, is not generally recognized.

Then, a lower level of circulating blood cells sometimes accompanies such conditions as leukemia, Gaucher's disease, Hodgkin's disease and reticulum cell sarcoma. In these disorders bone marrow replacement may be partly responsible at times but this is not often the case. In all these conditions greater stagnation of blood in the spleen might be expected to occur and all are conditions affecting the reticuloendothelial system. The actual mechanism whereby blood cell disintegration is accelerated in the spleen is not as yet established but the evidence at least points away from phagocytosis as any more than a passive secondary phenomenon.

DR. TURNER: With this background we may now turn to more detailed consideration of some of the clinical aspects of hypersplenism. Dr. Hyman will begin by discussing thrombocytopenic purpura.

DR. GEORGE A. HYMAN: I shall deal mainly with so-called idiopathic thrombocytopenic purpura. All other thrombocytopenic purpuras may be classified as secondary or "symptomatic" and fall under the following broad headings as suggested by Wintrobe: (1) chemical and physical agents; (2) blood disorders such as congestive splenomegaly (Banti's syndrome), Boeck's sarcoid, tuberculosis, Gaucher's disease of the spleen, disseminated lupus erythematosus, Felty's syndrome, in addition to leukemias and lymphomas; (3) infections.

The earliest suggestion of the role of the platelet in thrombus formation and blood coagulation was made by Bizzozero in 1882. A year later Krauss described a decrease in the number of platelets in a case of thrombocytopenic purpura. At the present time the popular view holds that platelets are produced by the megakaryocytes, have a life span of perhaps three to eight days and an ill defined characteristic of "stickiness." They clump at the site of blood vessel damage, plug the opening mechanically and form the basis of a clot. They then undergo lysis and release a vasoconstrictor substance which aids in hemostasis. At the same time the platelets take part in the activation of prothrombin and the formation of thrombin. Much experimental work has been done in an attempt to produce thrombocytopenic purpura in animals, especially with splenic extracts. The interpretation of results remains highly controversial and will not be discussed now.

The varieties of mode of onset and the clinical manifestations of the bleeding diathesis in idiopathic thrombocytopenic purpura are well known, as is its peak of incidence in childhood and early adult life. The physical findings depend on the site and extent of bleeding with petechial and purpuric lesions especially prominent. The spleen is rarely palpable and significant enlargement should cast doubt on the diagnosis.

Depression of the platelet count below 60,000 per cubic millimeter (direct method) is characteristic. The bleeding time is usually prolonged well above the accepted normal limit of three minutes and a positive tourniquet test, increased capillary fragility and failure of clot retraction are frequently found. Clotting time and prothrombin time (1-stage Quick) are normal. More than slight anemia is uncommon and when not explained in terms of blood loss suggests secondary thrombocytopenic purpura. Except for the occasional leukocytosis seen with severe hemorrhage the white count is normal.

Bone marrow aspiration is essential and reveals normal or increased activity. According to some authors, including Dameshek and Schwartz, there is an absolute increase in megakaryocytes, which frequently show diminished granularity and platelet formation. Eosinophils may be increased in number, and this is considered by some to indicate that a good response may be expected from splenectomy. In this clinic abnormalities of the megakaryocyte have not been found to correlate well with the type of purpura. We share the view that the primary purpose of bone marrow examination is to rule out leukemia and allied disorders of the marrow before proceeding with surgery.

In the milder form of the disease and in childhood, time, rest and patient observation are usually the initial treatment of choice. Transfusion should in general be reserved for red cell replacement as it possesses no other evident specific benefit. The platelet count remains un-

affected. A stimulating piece of research is under way in Cohn's laboratory at Harvard where success has been achieved in fractionating and concentrating the cellular components of the blood, including platelets. It remains to be seen what may be accomplished therapeutically with such preparations.

The first splenectomy for the condition was not performed until 1916 when Kaznelson, then a medical student, suggested this as a therapeutic measure on the hypothesis that accelerated phagocytosis of platelets takes place in the spleen. Splenectomy for idiopathic thrombocytopenic purpura was introduced at the Presbyterian Hospital by Dr. A. O. Whipple in 1924. Since then sixty-eight operations have been performed on patients eleven to fifty-four years of age, and 72 per cent have been followed up for four years or longer. Among them were fifty-one females and seventeen males, the usual 3:1 ratio. In forty-nine cases or 72 per cent it was judged that unequivocal benefit followed operation while the results were disappointing in nineteen or 28 per cent. The most successful results appeared in the age group under thirty. These are just the reverse of the results obtained in twenty-five patients treated conservatively, for the disease persisted in 72 per cent.

The absence of any immediate surgical mortality in this series is a direct tribute to skillful surgical technic and advances in methods of anesthesia and operative support. The experience in this hospital compares quite favorably with a reported countrywide operative mortality of 12 per cent and indicates that removal of the spleen in this condition is no longer as dangerous a procedure as was once believed. In most cases bleeding may cease in a matter of minutes after ligation of the splenic pedicle although platelet counts may take a period of a few hours to a few days to rise to normal. In addition, Elliott demonstrated that a postoperative decrease in capillary fragility may precede the rise in platelets, further suggesting multiple factors in the hemorrhagic tendency in this disease.

In the operated group there were five late deaths, eight months to nine years following operation, due to recurrence of purpura. In no instance of failure was an accessory spleen demonstrated although accessory spleens were found in 25 per cent of the sixty-eight patients operated upon, a figure corresponding closely with those of Curtis, Coller and other authors. This does not alter the advisability of reoperating upon certain carefully selected patients but emphasizes the need of careful initial exploration. Three of the patients not operated upon died, yielding an over-all mortality of 8.6 per cent of ninety-three cases. Seven of the eight deaths were due to cerebral hemorrhage and there were five other non-fatal instances of this complication, giving a total incidence of 12.9 per cent. In several instances surgery was contemplated but not undertaken because the true severity of the disease was not appreciated. Thus the indications for splenectomy in idiopathic thrombocytopenic purpura are (1) severe or locally dangerous bleeding uncontrolled by transfusion and imperiling life and (2) a recurrent or persistent chronic hemorrhagic tendency.

In secondary or "symptomatic" thrombopenic purpura splenectomy generally has been considered as contraindicated. However, benefit was observed in six of ten such patients operated upon at this hospital. The six comprised two cases of disseminated lupus erythematosus, three of Boeck's sarcoid and one secondary to gold toxicity. The four failures consisted of two patients with tuberculosis and two with gold toxicity. Again there were no operative deaths. It is of interest that splenectomy arrested purpura in five additional patients with disseminated lupus erythematosus reported in the literature as undergoing the operation, although the systemic disease remained unaffected. Obviously splenectomy is not indicated as routine treatment in secondary thrombocytopenic purpura; many instances are either self-limited or are associated with rapidly fatal diseases. It may be of benefit, however, in carefully selected cases.

A discussion cannot be concluded in this adrenal-conscious age without a word about those partners in therapy, ACTH and cortisone. These substances have been found useful in controlling the hemorrhagic manifestations of aplastic anemia, disseminated lupus erythematosus, thrombocytopenic purpura and leukemia, although platelet counts may not change. Customary doses seem to be followed by diminished bleeding on about the fifth day. A relapse is likely to be seen within a few days after treatment is withdrawn. However, reports now appearing in the literature suggest that a more prolonged remission of idiopathic thrombocytopenic purpura may be produced by ACTH or cortisone. Of course, follow-up has been short

and great caution must be exercised in final interpretation.

Two patients recently treated in this hospital illustrate these points. The first was a twenty-two year old dental technician who was seen in the hematology clinic complaining of easy bruisability and menorrhagia for six months. Epistaxes had occurred for several days. She had used a hair dye containing paraphenylenediamine on one occasion prior to onset of symptoms. On examination a few ecchymoses were noted on the lower legs, arms and mid-abdomen; the spleen was not palpable. The platelet count was 22,000 per cu. mm., bleeding time (Duke) 9½ minutes, hemoglobin 11.4 gm. per cent, red cell count 3,800,000 per cu. mm. and white cell count 7,000 per cu. mm. Cortisone was given intramuscularly in a course totalling 1.7 gm. over an eleven-day period. The response to therapy with a rise in platelets and disappearance of hemorrhagic phenomena after five days is shown in Figure 3. At present platelet counts have again fallen, approximating 30,000 per cu. mm., and the patient is showing easy bruisability again.

The second patient was a six and a half year old schoolboy admitted to Babies' Hospital with severe ecchymoses and epistaxes for thirty-six hours, and hematemesis and melena for twelve hours. Past and family histories were negative. On admission the patient had a temperature of 101°F., was pale and acutely ill with generalized purpura. There was continual bleeding from the gums, tongue and pharynx, and from the urinary and gastrointestinal tracts. The platelet count varied from 30,000 to 60,000 per cu. mm., bleeding time (Duke) was 20 minutes, hemoglobin 8.6 gm. per cent and white cell count 13,200 per cu. mm. with 76 per cent neutrophils. In view of the steadily downhill course in spite of four transfusions ACTH therapy was started on the fifth day of hospitalization. A total of 340 mg. was administered intramuscularly during a nine-day period. A rising platelet count and absence of further hemorrhagic phenomena were noted on the fifth day and this improvement has persisted to date. The therapeutic response can be seen in Figure 4.

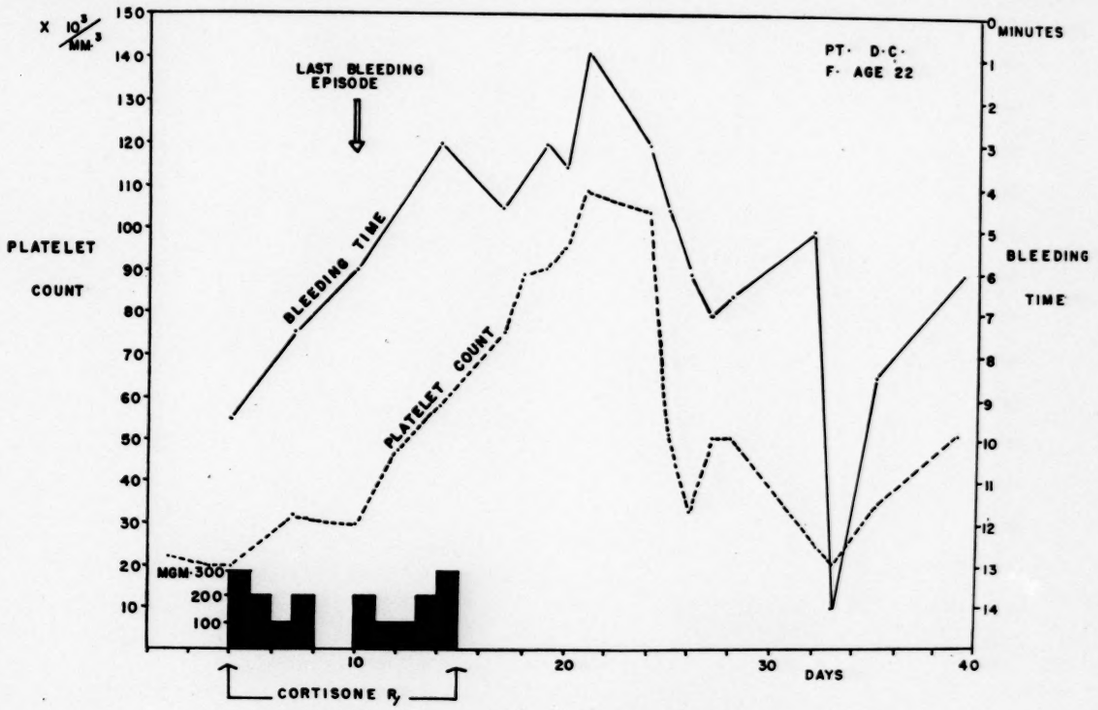

FIG. 3. Cortisone therapy in a case of thrombocytopenic purpura.

The suggestive effects obtained with these newer agents is quite provocative, especially since the mechanism of action is entirely unknown. Although this mode of therapy with ACTH or cortisone looks promising, it remains for time to indicate the true role these agents will play in the management of patients with

thrombocytopenic purpura and to what extent they may supplant splenectomy.

DR. TURNER: A particularly interesting syndrome which must be included in any current discussion of hypersplenism is primary splenic neutropenia. Dr. Ranney will discuss this subject for us and also consider in some detail the possible role of hypersplenism in the acquired hemolytic anemias.

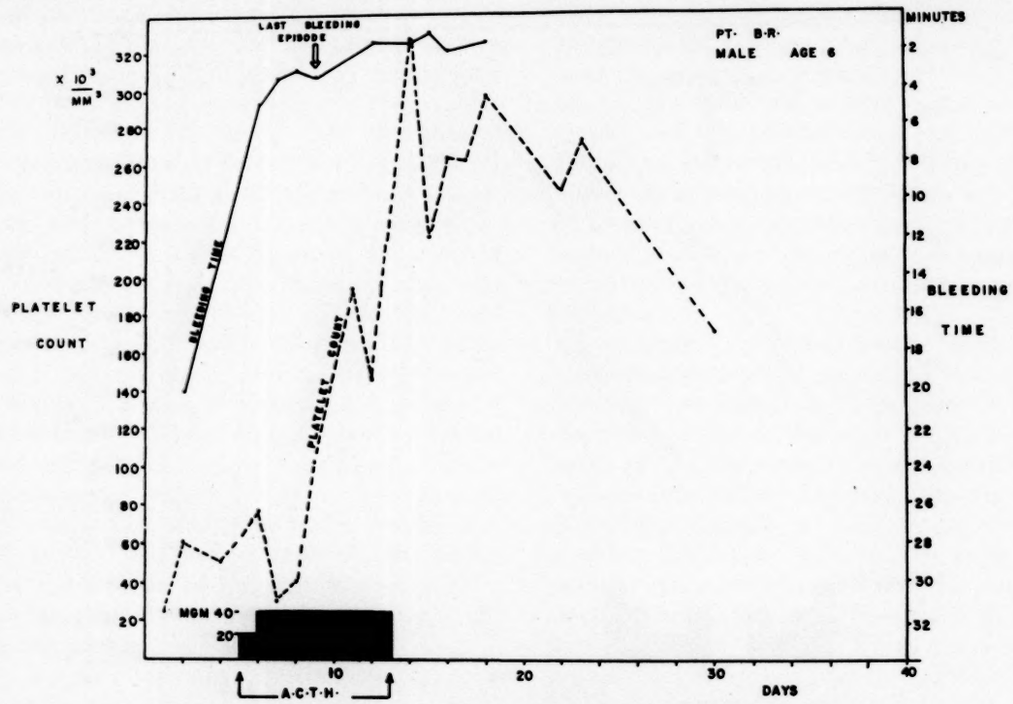

FIG. 4. ACTH therapy in a case of thrombocytopenic purpura.

DR. HELEN RANNEY: Of the syndromes which are today being considered as manifestations of hypersplenism perhaps the most unusual and certainly the most recently recognized is primary splenic neutropenia. In 1939 Wiseman and Doan reported several cases of a primary granulopenic syndrome with the following characteristics: (1) myeloid hyperplasia of normal cells in the bone marrow, (2) splenomegaly, (3) peripheral granulopenia and (4) therapeutic response to splenectomy. During the ensuing eleven years several more cases have been reported. The presence and duration of symptoms have been variable; some patients have had recurrent infections while others have not been particularly symptomatic. Spontaneous remission is uncommon if it occurs at all. Remission following splenectomy is prompt and sustained.

Figure 5 illustrates the course of a patient with primary splenic neutropenia. This patient had a reduction of circulating platelets in addition to neutropenia but had no clinical purpura. Similar reduction of platelets has been noted in other reported patients with this syndrome. Some of the patients have had anemia but in this patient the hemoglobin concentration and red blood cell count were normal throughout his course. The rise in neutrophils was noted a few hours after splenectomy; the rise in platelets occurred during the first week after operation. The patient has been clinically well with a normal blood count since operation over two years ago.

It should be noted that if splenomegaly is lacking splenectomy should be considered with caution; it may be extremely difficult to eliminate exposure to chemicals or ingestion of drugs as the etiology of the granulopenia.

Turning now to the acquired hemolytic anemias, among the most frequently encountered in this area is that known variously as chronic hemolytic jaundice, chronic spherocytic anemia or acholuric jaundice. Although this portion of the discussion is concerned with acquired hemolytic anemias, it is impossible to discuss the acquired without reference to the perhaps more familiar congenital hemolytic anemia. Both types have many clinical and hematologic characteristics in common: (1) Usually there is

anemia, varying in degree from mild to severe. (2) Both may have mild icterus or at least hyperbilirubinemia with increased fecal urobilinogen excretion. (3) The spleen is enlarged and usually palpable. (4) Reticulocytosis, increased osmotic and mechanical fragility and spherocytic microcytes may occur in both forms of the disease.

In the first decade of this century a congenital and an acquired form of the disease were differentiated clinically. The acquired form of the disease occurred without a family history of a similar disorder; patients with acquired hemolytic anemia had the onset of the disease later in life and were frequently sicker and more anemic than patients with congenital hemolytic anemia. The spherical microcytes so frequently found in the congenital disorder were often absent in the acquired form. Obviously none of these criteria necessarily provided a definitive differentiation of the two forms of the disease, particularly when in rare cases patients with a family history of hemolytic anemia were found to have the onset of the disease in late adult life.

During the next twenty years this division into acquired and congenital hemolytic jaundice was almost entirely superseded by classifications based on the histology of the spleen, on the presence of spherocytes or on response to splenectomy. The presence of a coexisting disease, usually lymphoma or leukemia, with splenomegaly and hemolytic anemia was occasionally noted and some workers considered all chronic hemolytic anemias of other than the typical familial variety to be secondary to other disease.

However, during the last decade, on the basis of two new diagnostic criteria, chronic hemolytic anemia has again been divided into an acquired and a congenital form. The acquired form is further subdivided into a primary or idiopathic form and a secondary form in which there is a coexisting disease. These new tests are the serologic or Coombs test and the determination of the survival time of transfused erythrocytes.

The Coombs test is believed to detect the presence of a coating antibody on the surface of the red blood cells. Coombs' antiserum used for this test is produced in rabbits by the repeated injection of human globulin. This rabbit antihuman globulin will agglutinate previously unagglutinated saline suspensions of human erythrocytes coated with antibodies. For the direct Coombs test a 2 per cent suspension of the patients' erythrocytes is washed several times with saline; one or two drops of rabbit antihuman globulin are then added. The presence of agglutination indicates the adsorption on the erythrocytes of an antibody which was not removed by washing with saline. The direct Coombs test is usually positive in acquired

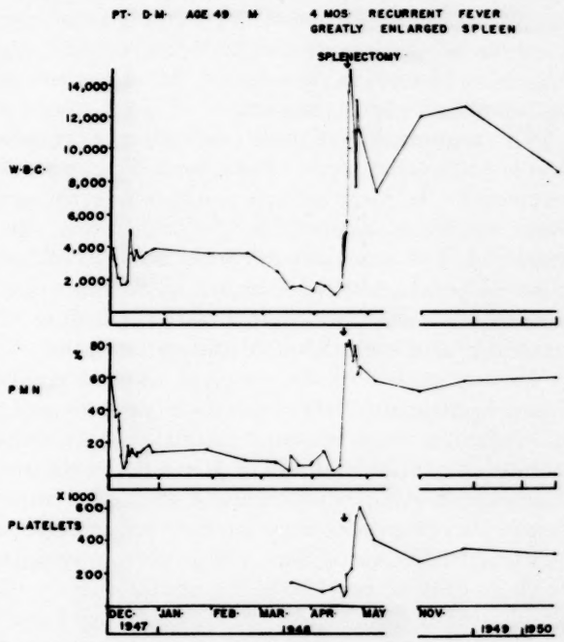

FIG. 5. A case of splenic neutropenia.

hemolytic anemia and negative in the congenital type of the disease. The presence of a positive direct Coombs test implies, of course, that these individuals have produced antibodies which coat their own red blood cells. The antigenic stimulus for this antibody formation is completely unknown.

For the indirect Coombs test the patient's serum is incubated with normal compatible erythrocytes. After these cells are washed with saline the direct test is applied. Here the test is used to detect the same kind of antibody in the patient's serum by adsorbing it on normal cells. The indirect Coombs test is positive in a minority of cases of acquired hemolytic anemia.

Other serologic technics may be used to demonstrate auto- and iso-agglutinins as well as hemolysins in the sera of patients with acquired hemolytic jaundice. The agglutinins can frequently be demonstrated if bovine albumin is used as a diluent. In addition, reactions employing the acidified sera of these patients have been described recently. If the serum of a patient with acquired hemolytic anemia is acidified to

a pH of 6.5 to 6.7 and incubated with normal homologous erythrocytes, agglutination usually occurs. If the pH of this suspension is subsequently raised to 8.0, the agglutination disappears but the red cells can be agglutinated once more by the addition of Coombs' antihuman globulin. A hemolysin, active at pH 6.8 to 7.0 in producing sensitization or hemolysis of the patients or of normal compatible erythrocytes, has been found in the serum of one case of acquired hemolytic anemia.

The relationship of these serologic abnormalities to each other is not established. They do not necessarily occur together and one serologic reaction may diminish while another is unchanged. For practical purposes the use of the direct Coombs test represents a most important contribution to the clinical differentiation of acquired and congenital hemolytic anemia.

Determination of the survival time of transfused erythrocytes has yielded the second point of differentiation between acquired and congenital hemolytic jaundice. If normal erythrocytes are transfused to a patient with congenital hemolytic jaundice, they survive for a normal period of 120 days; if they are given to a patient with acquired hemolytic jaundice, their survival time is diminished. If, on the other hand, red blood cells from a patient with congenital hemolytic jaundice are given to a normal individual, the survival time of these erythrocytes is shortened; if the donor is a patient with acquired hemolytic jaundice and the recipient normal, the survival time of the transfused erythrocytes will be normal. Thus the patient with acquired hemolytic jaundice exhibits these two abnormalities: (1) a positive Coombs test indicating the presence of a coating antibody on his erythrocytes and (2) rapid destruction of normal red cells which he receives by transfusion. The fact that Coombs-positive red blood cells from patients with acquired hemolytic jaundice survive for a normal period of time when transfused to normal subjects indicates that the mere presence of this antibody does not necessarily doom these erythrocytes to premature destruction. The complete mechanism of the shortened erythrocyte life span in these patients is not known.

Another difference between acquired and congenital hemolytic anemia is the presence of methemalbumin in the plasma of patients with the acquired form of the disease. This substance is present in only very small amounts or absent in the serum of patients with congenital hemolytic anemia. This test has not been nearly as extensively evaluated as the serologic test and cross transfusion experiments, and at this time its validity and significance are not entirely clear. With extensive intravascular hemolysis such as occurs in blackwater fever or transfusion reactions large amounts of hemoglobin may be released into the circulation. Here a large fraction of the hemoglobin is excreted in the urine but some of the hemoglobin is split to form hematin and globin. Hematin thus released into the circulation combines with circulating plasma albumin to form methemalbumin, which may be identified by spectroscopic methods.

Whether the presence of methemalbumin in acquired hemolytic jaundice, and its absence in congenital hemolytic jaundice, reflects a difference in the rate of red blood cell destruction in the two diseases is not known. Certainly the anemia may be more severe and the erythrocyte destruction more accelerated in the acquired disorder than in most cases of the congenital type, and no study of the correlation of the test for methemalbumin with the excretion of fecal urobilinogen has been published. The presence of circulating methemalbumin has been interpreted as indicating intravascular hemolysis but any interpretation is speculative at the present time.

In part this discussion of classification of chronic hemolytic anemias has been based upon the irregularity of response of patients with acquired hemolytic anemia to splenectomy. It must be admitted that while the serologic and erythrocyte survival studies have added to our understanding of the disease they have not necessarily enhanced our ability to prophesy the effects of removal of the spleen. A few cases, and again I do not know how they can be distinguished at onset, recover spontaneously and this adds to the difficulties in assessment of the operative results. Although nearly 100 per cent of patients with congenital hemolytic jaundice are benefited by splenectomy, only 50 to 60 per cent of patients with the acquired disorder are benefited. In the patients with acquired hemolytic anemia who respond to splenic extirpation the response in terms of red blood cell rise is usually slower than in the congenital disorder and the positive Coombs test may persist for months or years after the operation. Postoperatively the elimination of normal erythrocytes transfused to the patient is usually slower than

before operation but the survival time does not necessarily return completely to normal despite clinical recovery.

It has already been stated that some workers believe all cases of acquired hemolytic anemia to be secondary to another coexisting disease, whether or not this disease can be identified. We have preferred to classify cases without other demonstrable disease as idiopathic. The same hematologic syndrome may occur in the presence of another disease, notably lymphoma or leukemia. The secondary type exhibits the same abnormalities as the idiopathic form: (1) positive Coombs' test and (2) shortened survival time of normal transfused erythrocytes. Twenty-one cases of acquired hemolytic anemia associated with leukemia were reported recently; all showed a diminished red cell survival time. In five of these patients splenectomy was performed following which there was clinical improvement and the life span of transfused erythrocytes returned to normal. Splenectomy is considered in cases of leukemia with a chronic hemolytic type of anemia in the presence of the following findings: (1) splenomegaly, (2) erythroid hyperplasia of the bone marrow and (3) absence of marked myeloblastic (or lymphoblastic) proliferation in the bone marrow. In the secondary form of the disease considerations of splenectomy should obviously include the ultimate prognosis of the coexisting disease. Good clinical judgment is necessary in the selection of patients for splenectomy.

For the sake of clarity the serologic and erythrocyte survival studies have been presented as clear-cut and definitive differences between acquired and congenital hemolytic anemia. Actually, a few cases of chronic hemolytic anemia with a family history of the disorder but with positive serologic tests have been described and at least one case of familial hemolytic anemia and increased destruction of normal transfused erythrocytes has been reported. In addition the Coombs test is positive in patients who have anti-Rh blocking antibody and may be positive, in the absence of any manifestations of a hemolytic type of anemia, in patients with disseminated lupus erythematosus or periarteritis nodosa.

The effects of ACTH and cortisone on chronic hemolytic anemias may be dramatic; evidence of increased hemolysis may diminish or cease. However, sustained remission after these drugs are discontinued usually does not occur. The mechanism of this activity of these hormones is not known.

STUDENT: Do studies of the red cell life span have any practical value? Do they help to decide whether splenectomy should be performed?

DR. RANNEY: Usually an erythrocyte life span study is not necessary to establish the presence of hemolytic anemia. In occasional cases, particularly of secondary acquired hemolytic anemia, a markedly shortened red blood cell life span may be taken as a consideration in favor of splenectomy. Of course, increased red blood cell destruction occurs in a variety of clinical syndromes, e.g., sickle cell anemia, favism and Cooley's anemia, which are not usually benefited by splenectomy.

STUDENT: You have not said anything about the epinephrine test for hypersplenism. I believe this may be useful in the diagnosis of hypersplenic states.

DR. TURNER: It is claimed that the epinephrine test contributes evidence on the score of whether the spleen is sequestering red cells or granulocytes or platelets. The test consists in taking note of changes in the numbers of these formed elements after subcutaneous injection of 0.5 to 1.0 ml. of 1:1000 adrenalin. Thus in idiopathic thrombocytopenic purpura such administration of adrenalin may be followed by transitory increases in platelet count without corresponding increments of red and white cells. This sort of change has been interpreted as signifying the release of sequestered platelets from the spleen.

It may be so. There can be no doubt that epinephrine may contract the spleen and must then squeeze cells into the splenic vein. Still epinephrine has widespread and complex effects and may bring about increments of formed elements in unknown ways and in an irregular fashion. I believe it is fair to say that there are differences of opinion about the value of the epinephrine test in establishing a diagnosis in the diseases under consideration. My own view is that the results of the test should not weigh heavily in arriving at a decision in a given clinical problem.

SUMMARY

DR. TURNER: It will by now have become apparent that our inclination is to avoid generalizations about the relation of the spleen to the various diseases under discussion. It appears that in congenital hemolytic jaundice

and in many instances of acquired hemolytic jaundice the red cells are abnormal and are consequently trapped by the peculiar splenic circulation. Such stasis accelerates disintegration of the cells but there is not agreement that this process involves increased phagocytosis by the reticuloendothelial cells rather than increased intravascular hemolysis. Splenectomy is an effective remedy for congenital hemolytic jaundice because it eliminates the paramount site of erythrostasis. By the same token it may alter the course of acquired hemolytic jaundice but cannot otherwise influence the final outcome if the accelerated blood destruction is merely a manifestation of a disease such as lymphoma.

The findings in idiopathic thrombocytopenic purpura are different. The spleen is not enlarged; there are no regular histologic features to distinguish it from the normal organ. The hypothesis of some hormonal function of the spleen has been advanced to account for the fact that splenectomy is of benefit in this disease. No such hormone has been demonstrated and no cells peculiar to the spleen that might elaborate a special hormone have been identified. It is entirely possible, however, that any substances elaborated by the lymphocyte or macrophage systems would have effects that are magnified to the point of recognition only in the presence of the spleen, because in that organ the formed elements of the blood run a sluggish course and come into most intimate contact with the reticuloendothelial system. Note must be taken of a very recent report that constitutes a major contribution to the problem of purpura: Harrington and co-workers at St. Louis have shown that if the blood of patients with idiopathic thrombopenic purpura is transfused into non-thrombopenic human subjects it may induce thrombopenia and even purpura. The factor responsible appears to reside in the plasma. This important discovery affords a most promising line of investigation of the problems under discussion and one that may soon lay at rest some of the speculations hitherto entertained.

Splenectomy remains the treatment of choice for chronic or severe idiopathic purpura of the thrombocytopenic type. ACTH and cortisone therapy are being tried; our own limited experience with these agents in adult purpura has been that they may be expected to bring about partial or almost complete remissions of purpura but that these remissions are likely to last only so long as the hormones are administered.

Primary splenic neutropenia is a rare disorder which appears to respond well to splenectomy.

Chronic portal hypertension characterizes virtually all examples of what has been called Banti's syndrome or congestive splenomegaly with hematocytopenia. Stagnation of blood takes place in closed channels in the spleen while there is an increase in number of reticulum cells. A humoral mechanism may be concerned in the blood picture but this is not proved. The therapeutic management of this problem is difficult and a discussion of it would have to go beyond the limits set for this clinic. Insofar as the spleen is concerned it would appear that splenectomy is often followed by a movement of the numbers of formed elements of the blood toward normal levels. However, this simple operative procedure is now usually accompanied with some sort of portocaval shunt inasmuch as the most important immediate part of the clinical problem is likely to be bleeding from esophageal varices.

Clinico-pathologic Conference

Aortic Insufficiency with Cardiac Failure and Recurrent Abdominal Pain

STENOGRAPHIC reports, edited by Robert J. Glaser, M.D. and David E. Smith, M.D., of weekly clinico-pathologic conferences held in the Barnes Hospital, are published in each issue of the Journal. These conferences are participated in jointly by members of the Departments of Internal Medicine and Pathology of the Washington University School of Medicine and by Junior and Senior medical students.

THE patient, E. T. (No. 134196), a white married farmer sixty-three years of age, was admitted to the Barnes Hospital for the first time on April 6, 1946, complaining of abdominal pain, shortness of breath and swelling of the ankles. The family history was irrelevant. The patient had enjoyed excellent health until the onset of his present illness. Systemic review was non-contributory.

Six years prior to admission the patient noted for the first time slight dizziness. He consulted his physician who told him that his blood pressure was elevated; the patient was advised to moderate his activities. Subsequently, he did only light work and felt entirely well until five to six months prior to entry when he developed abdominal pain localized to the upper portion of his abdomen. The pain was intermittent, unrelated to meals and unassociated with any other digestive disturbances. Pressure over the left upper abdomen accentuated the pain. Although at no time was the distress severe, it increased in frequency until it occurred almost daily. Four months before entry the patient noted dyspnea on exertion which was rapidly progressive; soon he was short of breath even at rest and his ankles swelled increasingly. At the time of entry both dyspnea and edema were severe.

Physical examination on admission revealed the temperature to be 38°C., pulse 96, respirations 24 and blood pressure 180/90. The patient appeared chronically ill and was moderately orthopneic. His complexion was sallow and there was slight cyanosis of the lips and nail beds. The pupils reacted normally to light and accommodation. Examination of the fundi revealed arteriolar narrowing and distention of the veins. No hemorrhages or exudates were seen. Examination of the upper respiratory tract was negative. The trachea was in the midline and there was no tracheal tug. The neck veins were slightly distended with the patient sitting up at a 45 degree angle. Examination of the lungs revealed flatness to percussion and absent breath sounds at the right base, extending up to the angle of the scapula; no rales were heard. The border of cardiac dullness was percussed 12 cm. to the left of the mid-sternal line in the sixth interspace. There were no thrills. A grade I systolic murmur was heard at the aortic area, and a grade III high pitched blowing diastolic murmur was audible at the base; it was loudest in the third and fourth interspaces just to the left of the sternum. An apical diastolic murmur was not present. The peripheral arteries were thickened and the radial pulses were collapsing in type. Abdominal examination revealed a large ventral hernia. The liver edge was felt 3 cm. below the right costal margin and was distinctly tender. Signs of fluid were present. There was 4+ pitting edema over the lower extremities and 2+ pitting edema over the sacrum. The neurologic examination was within normal limits.

Laboratory data were as follows: Blood count: red cells, 4,950,000; hemoglobin, 15 gm.; white cells, 11,000 with 4 per cent stab forms and 73 per cent segmented forms. Urinalysis: specific gravity 1.018; albumin 4+; sugar, negative; sediment, many granular casts. Blood Kahn test, negative. Venous pressure: 230 mm. of water. Circulation time (Decholin): 50 seconds. Blood chemistry: non-protein nitrogen, 23 mg. per cent; total proteins, 5.9 gm. per cent; albumin, 3.4 gm. per cent; globulin, 2.5 gm. per cent; CO_2 combining power, 33.4 mEq./L.; chlorides, 90 mEq./L.; icterus index, 13.4 units; cephalin-cholesterol flocculation test, negative. Roentgenogram of the chest: The cardiac silhouette was enlarged to the left and to the right and the

aorta was lengthened. There was infiltration in the perihilar region on both sides and a collection of fluid was noted in the right pleural cavity. Electrocardiogram: inverted T waves in lead I, diphasic T waves in lead II; S-T segments slightly depressed in leads I, II and CF_4. Impression: myocardial damage or digitalis effect.

On the night of entry the patient was given aminophyllin intravenously because of Cheyne-Stokes respirations. A thoracentesis was performed; 750 cc. of clear fluid were obtained which had the characteristics of a transudate. Following these measures and the institution of digitalis therapy, the patient's dyspnea decreased and he felt much improved. A brisk diuresis occurred and in the first several days of hospitalization the patient lost 12 pounds. Concomitantly, the edema decreased. During the first hospital week his temperature ranged between 37° and 38°C. The albuminuria and casts disappeared but the white blood cell counts continued to range between 14,000 and 19,000. The patient persistently complained of pain in the abdomen which he described as a severe "ache," particularly in the right upper quadrant. On physical examination no tenderness or muscle spasm could be made out. A flat film of the abdomen was unrevealing aside from hypertrophic osteoarthritic spurs on the lumbar vertebrae.

In an attempt to elucidate further the nature of the pain, cholecystograms were taken; there was only faint visualization of the gallbladder throughout the series and a diagnosis of diseased gallbladder was made. An open film of the urinary tract failed to show distinct kidney shadows. Intravenous pyelography was performed and only a small amount of the dye appeared in the pelvis or calices of either kidney.

At the end of the first hospital week the patient became slightly jaundiced. His temperature was 38°C.; the white cell count was 19,600 with a marked left shift in the differential. In the next few days the patient continued to be free of signs or symptoms of cardiac decompensation but icterus persisted. He complained less of abdominal discomfort; his pain was relieved by codeine. At no time was the abdomen distended, but the patient developed some tenderness and muscle spasm in the right upper quadrant. He was seen by a surgical consultant who did not believe that the findings were due to primary inflammatory disease of the abdomen.

During the course of the second hospital week the patient's icterus index fell to 6.7 units. Another thoracentesis was performed but only 250 cc. of faintly pink, slightly cloudy fluid were obtained. The specific gravity of the fluid was 1.019, and the cell count revealed 192 cells with acid of which 95 per cent were mononuclear forms. The protein content was 2.4 gm. per cent. Smears and cultures revealed no pathogens.

A repeat chest film showed only a decrease in the heart size, and a second electrocardiogram showed no changes from those recorded on admission. During the remainder of his hospital course the patient continued to have mild fever, a slight leukocytosis and a moderate anemia, but at the time of discharge on May 2, 1946, his abdominal pain had decreased markedly and there were no signs of cardiac decompensation.

After leaving the hospital the patient took digitalis and received occasional mercurial diuretics. On the whole he did rather well; he remained free of significant symptoms or signs of heart failure. Ten days before his second admission he suddenly lost consciousness and developed a left hemiparesis. He was admitted to an outside hospital where, because he was unable to void, an indwelling catheter was inserted. Subsequently the patient passed bloody urine for several days. Hematuria then subsided and the catheter was removed. Once again retention occurred and difficulty was encountered in reinserting the catheter. The patient's non-protein nitrogen rose to 65 mg. per cent, and he was transferred to the Barnes Hospital on July 30, 1950. During the four years since his first admission the non-protein nitrogen had averaged 40 mg. per cent on several determinations.

Physical examination at the time of entry revealed the temperature to be 38.5°C., pulse 100, respirations 48 and blood pressure 170/70. The patient appeared well developed but somewhat dehydrated. He was unconscious and unresponsive to painful stimuli. Respirations were loud and of the Cheyne-Stokes type. The skin was cold and moist. The pupils were constricted and the eye grounds were not well visualized. A few rales were heard at both lung bases. Examination of the heart revealed cardiac dullness to extend 7 cm. to the left of the midsternal line in the fifth interspace. The sounds were of poor quality and obscured by stertorous breathing. The rhythm was totally irregular. Diastolic and systolic murmurs were again heard over the aortic area and along the left

sternal border. The peripheral pulses were collapsing in character. Abdominal examination was negative; a limited neurologic examination revealed hyperactive reflexes on the left side, but the patient was able to move both arms and legs.

The laboratory findings included the following: Blood count: red cells, 4,120,000; hemoglobulin, 12.3 gm.; white cells, 19,400; differential count: stab forms 2 per cent, segmented forms 94 per cent, lymphocytes 3 per cent, monocytes 1 per cent. Urinalysis: specific gravity, 1.005; albumin, 2+; sugar, negative; sediment, numerous hyalin and granular casts and many red and white blood cells per high power field. Blood chemistry: non-protein nitrogen, 47 mg. per cent; chlorides, 110 mEq./L.; CO_2 combining power, 17.5 mEq/L.; blood sugar, 191 mg. per cent.

Soon after entry the Cheyne-Stokes respirations disappeared and the patient exhibited rapid, shallow, regular breathing at a rate of 40 per minute. An indwelling catheter was inserted without difficulty and the patient's urinary output was quite adequate. On the second hospital day he developed large confluent, purplish red areas over the abdomen; the abdomen became rather tender and rigid, and a questionable fluid wave was elicited. The bowel sounds gradually disappeared. Continuous suction with a Wangensteen tube was instituted. The patient was given intramuscular penicillin and streptomycin and intravenous aureomycin. The ecchymotic areas spread to involve the inner aspects of both thighs down to the knees.

On the third hospital day the patient's temperature was 39.4°C., pulse 112, respirations 52 and blood pressure 110/50. He continued to be obtunded. The ecchymotic areas extended to the chest. The rales which had been heard at the time of admission at the lung bases became more numerous; the abdominal signs were unchanged. The non-protein nitrogen rose to 101 mg. per cent. The amylase was 250 units. Blood cultures taken on the first two days of hospitalization remained sterile. The patient expired August 1, 1950, without ever regaining a conscious state.

CLINICAL DISCUSSION

DR. HARRY L. ALEXANDER: This case presents several interesting problems. At the outset it might be well to consider the cardiovascular aspects of the case. The patient was a farmer who had enjoyed good health most of his life. When he was fifty-seven he was told that he had hypertension and subsequently he limited his activity quite sharply. Six years later he developed severe cardiac decompensation rather rapidly. Dr. Massie, would you care to suggest the etiology of the patient's heart disease.

DR. EDWARD MASSIE: It seems to me that the most striking finding on cardiac examination was the loud diastolic murmur which obviously arose at the aortic area. At the time of the first admission the systolic murmur was not impressive. In considering the etiology of the patient's heart disease one must think first of the causes of aortic insufficiency. One is rheumatic heart disease, a second syphilitic heart disease, and a third arteriosclerotic heart disease with a diastolic aortic murmur arising on the basis of dilatation of the aortic ring. Statistically, arteriosclerotic heart disease as a cause of aortic insufficiency is much less likely than either rheumatic heart disease or syphilis; against it also is the fact that the patient's systolic murmur was not very striking. Usually when aortic insufficiency is due to arteriosclerosis, a systolic murmur suggestive of aortic stenosis is present. Without any particularly good evidence for so doing, I would favor rheumatic heart disease as the cause of the patient's valvular disease. The negative serologic test is against syphilis although it does not rule it out.

DR. ALEXANDER: Does the diastolic murmur of aortic insufficiency due to arteriosclerosis have a different character than that due to syphilis or rheumatic heart disease?

DR. MASSIE: No, I believe that the sound of the murmur is the same regardless of the nature of the pathologic change. The presence of a tambour aortic second sound, not described here, suggests syphilitic heart disease.

DR. ALEXANDER: What about the wide pulse pressure? You will recall that the patient had a Corrigan pulse.

DR. MASSIE: Aortic insufficiency, regardless of its etiology, may be manifested by the classical peripheral signs of increased pulse pressure. In a patient who also has aortic stenosis, one might not expect to find striking peripheral signs; I do not believe that this patient exhibited signs of aortic stenosis, and I am afraid that the presence of a wide pulse pressure is not particularly helpful in making an etiologic diagnosis in this particular instance.

DR. ALEXANDER: Dr. Smith, what is your feeling on this matter? Do you believe the patient had rheumatic heart disease?

DR. JOHN R. SMITH: Yes, that seems very likely. The negative blood Kahn test is of some help in ruling out syphilis, for in patients with syphilitic aortic valvulitis the serologic test for syphilis is usually positive.

DR. W. BARRY WOOD, JR.: Would Dr. Massie describe arteriosclerotic aortic insufficiency in a little more detail? Is it due to dilatation of the aortic ring from sclerosis at the base of the aorta?

DR. MASSIE: It is usually due to one of two causes; first, dilatation of the aortic ring accompanying a dilated aorta just distal to the valve; and second, as a concomittant of the stenotic change in calcific aortic stenosis.

DR. WOOD: Are the valves themselves normal in the first group?

DR. MASSIE: When aortic insufficiency is due to dilatation of the ring, the valves themselves are normal. In aortic stenosis, of course, the valves are the site of pathologic change. Whether the stenosis arises primarily on the basis of previous rheumatic infection or not is a much debated point. I have also noted aortic insufficiency occasionally in the presence of severe anemia.

DR. WOOD: In my experience aortic diastolic murmurs are rarely hemic in origin.

DR. CARL V. MOORE: I believe I have heard the murmur of aortic insufficiency, thought to have arisen purely on the basis of anemia, only twice.

DR. WOOD: In the presence of hypertension the occurrence of an aortic diastolic murmur is not especially rare in patients with normal valves. I was not aware, however, that the murmur of aortic insufficiency occurred on the basis of arteriosclerotic changes in the aorta in the absence of hypertension.

DR. MASSIE: I think your point is well taken; I do not recall having heard the murmur of aortic insufficiency in a patient who at postmortem had normal valves unless hypertension was also present. The patient under discussion today, for example, did have moderate hypertension.

DR. ALEXANDER: In the patient with aortic insufficiency due to hypertension, Dr. Wood, is it your understanding that the aorta may be widened but the valves are normal?

DR. WOOD: The valves are certainly normal when the pathologist examines them at the time of autopsy. The presence of the murmur is thought to depend on whether the ring is widened. Our usual explanation of aortic insufficiency due to hypertension is that the ring is widened and the cusps therefore do not close completely. Whether that is correct or not, I am not sure.

DR. ALEXANDER: Dr. Moore, would you comment on this particular point?

DR. ROBERT A. MOORE: If the valve ring is measured at autopsy, when there is of course no tone, one cannot demonstrate relative insufficiency. On the other hand, I am sure that in life, when the factor of structural tone is operable, relative insufficiency may exist in the presence of normal valves in hypertensive patients.

DR. ALEXANDER: To go on with the discussion, it will be remembered that the patient's heart failure responded well to the usual management, and he apparently had no further serious difficulty from heart failure. Four years later he suddenly developed hemiplegia which we assume arose on a vascular basis. The problem of degenerative vascular disease is a most important one in clinical medicine, and in the past year or two there has been much activity in laboratories studying the pathogenesis of arteriosclerosis. I have asked Dr. Schroeder, therefore, if he would comment on some of the newer studies.

DR. HENRY A. SCHROEDER: The etiology of arteriosclerosis is generally unknown. It is being recognized, however, as a disease; a very common disease, surely, but no longer is it considered a "normal" concomitant of the process of aging. Arteriosclerosis is relatively rare in races whose diets contain little fat, and it is common in countries where people eat a lot of dairy products. It also occurs in hyperlipemic conditions. We should differentiate between atherosclerosis, which is an intimal disease, and that condition which we loosely call arteriosclerosis and which is more or less the end stage of the intimal disease. One pathogenetic, if not etiologic factor in atherosclerosis, which we may accept tentatively, concerns the deposition of fats.

Two other factors, neither of which can be considered as definitely etiologic at the present time, may be mentioned. The first concerns pressure. It is a common observation that the higher the intravascular pressure, the more arteriosclerosis there is. The cholesterol content

of arterial wall increases with age but it also increases with pressure. Atherosclerosis seems more common when there are higher pressures; for example, in the pulmonary arteries in pulmonary hypertension; in the legs more than in the arms, where hydrostatic pressure may be the only difference between the two localities; in coarctation of the aorta proximal to the constriction; in occlusive disease of the aorta, above, not below the occlusion; in hypertension; in portions of the coronary arteries particularly in those of the left ventricle where the pressure during systole changes to a lateral pressure due to muscular contraction; and finally, even in the veins where there are arteriovenous fistulas. Therefore, the rate of deposition of lipids appears to be a function of pressure.

The second factor seems to be that of permeability. The nutrition of the subintimal area and possibly the media of the arterial wall seems to occur through diffusion, which is a function of the hydrostatic pressure of blood. The intimal lining apparently is a fatty layer because blood does not wet it but is capable of selective absorption. Hyperthyroidism, iodine and thiocyanates seem to affect the permeability of the intima. Perhaps this factor increases with age.

Theories as to the role of cholesterol are as numerous as the precursors and metabolites of cholesterol itself. Ever since cholesterol was discovered a hundred years ago it has been implicated in arteriosclerosis. In the early part of this century rabbits were first made atherosclerotic by feeding cholesterol to them. Other animals and chickens now suffer from this laboratory disease. The importance of the cholesterol level in the blood has been in and out of style. One may follow the cycle by watching the diets of those working on the problem; whether eggs are served for breakfast or not allowed in the house.

A group for the study of disease of the coronary arteries in Boston (Gertler, et al.) restudied the problem and found that the level of esters was high, but not of free cholesterol, in patients recovered from coronary occlusion.[1] There was a considerable overlap of "normal" levels and those of subjects believed to have arteriosclerosis as determined by the presence of coronary occlusion. This overlap has always led people to believe that cholesterol may not have much of a primary role. There is, however, no definitive diagnostic test for the presence of atherosclerosis other than the presence of its serious secondary pathologic changes. The evidence implicating cholesterol is as follows: it is in the lesions; we can produce the disease in experimental animals by feeding them cholesterol although very high blood levels must be maintained; the earliest lesions appear following injections of cholesterol into rabbits; and diseases with hypercholesterolemia dispose to atherosclerosis. Can one lower the blood levels? Cholesterol can be synthesized by the body from acetate; the total intake of fat influences the amount absorbed; apparently the diet must be almost completely fat-free to achieve a moderate reduction.

Lately the chylomicrons found in blood on dark field observation have been studied by some investigators. Chylomicrons are fat globules containing about 30 per cent cholesterol; they increase after a fat feeding. In older people the level increases much more and returns to normal much later than in younger people. Moreton believes that chylomicrons are taken by the lymphatics into subintimal tissues in a soluble form; after reabsorption of the fat the cholesterol fraction, a rather insoluble material, is the predominant residue.[2] This is engulfed by macrophages from reticulo-endothelial cells, leaving the foam cells which we see in relatively early stages.[3]

The ultracentrifuge has been used by Gofman et al. in California to float the fatty particles in serum.[4] By measuring the rate of rise or reverse sedimentation index, they have demonstrated specific particles in atherosclerotic sera with S_f (Svedberg flotation) values of 10 to 20, which are distinct from normal macromolecules of different values and from chylomicrons. These particles are lipoproteins. There is a high proportion of this type of particle in hyperlipemic states. They studied some 1,500 cases of hyperlipemia, hypothyroidism, hypertension, coronary insufficiency and nephrosis, and the correlation was excellent. The particles appeared in cho-

[1] Gertler, M. M., Garn, S. M. and Lerman, J. Inter-relationship of serum cholesterol, cholesterol esters and phospholipids in health and in coronary artery disease. *Circulation*, 2: 205, 1950.

[2] Moreton, J. R. Atherosclerosis and alimentary hyperlipemia. *Science*, 106: 190, 1947.

[3] Moreton, J. R. Physical state of lipids and foreign substances producing atherosclerosis. *Science*, 107: 371, 1948.

[4] Gofman, J. W., Jones, H. B., Lindgren, F. T., Lyon, T. P., Elliott, H. A. and Strisower, B. Blood lipids and human atherosclerosis. *Circulation*, 2: 161, 1950.

lesterol-induced experimental atherosclerosis. Furthermore, low fat diets decrease the number of these particular particles in man. If this work is substantiated, it may prove to be a diagnostic test of some specificity for the presence of atherosclerosis or predisposing conditions. In that event studies which now take decades to perform can be done in months, and the whole subject will be prone to investigative attack. At present there is probably little to be done in the way of therapy, except to eliminate fat from the diet completely, a most rigid regimen.

Atherosclerosis is probably the result of a general biochemical alteration in metabolism, one of the characteristics of which is an abnormality in the use of fats. Its development is influenced by physical, physiologic and chemical factors. Understanding of its pathogenesis is only beginning.

DR. ALEXANDER: Thank you very much Dr. Schroeder. Let us now consider the abdominal pain which was a very puzzling problem in this case. It began some five years before the patient's death and continued until his death. Dr. Rouse, I believe you saw the patient on his second admission.

DR. ERNEST T. ROUSE: That is correct, Dr. Alexander.

DR. ALEXANDER: What was your impression about the nature of the pain when you first saw him?

DR. ROUSE: When the patient was admitted the second time the abdominal pain was not the primary issue; he had been sedated heavily for the trip here and was unresponsive on arrival, and abdominal pain did not become apparent until the second day.

DR. ALEXANDER: Dr. Scheff, does the history suggest a cause for the pain which developed?

DR. HAROLD SCHEFF: Two definite possibilities come to mind. First, the patient may have had gallbladder disease with calculi; and second, it is also possible that he had recurrent pancreatitis. If he had pancreatitis, the terminal episode was probably hemorrhagic in type.

DR. ALEXANDER: Is it conceivable, that the patient had pancreatic disease which followed this course for five years?

DR. SCHEFF: Chronic relapsing pancreatitis, as described by the Mayo Clinic group, would fit this clinical picture.

DR. ALEXANDER: Dr. Rouse, when the abdomen became distended and bowel sounds disappeared, were you worried about intestinal obstruction?

DR. ROUSE: Yes, we were. In addition, we were impressed by the skin lesions which appeared over his abdomen.

DR. ALEXANDER: Were these purpuric spots?

DR. ROUSE: No, they were reticular in character, much as one sees on the extremities of a person in shock. This patient, however, was not in shock and the lesions appeared first over the abdominal wall and not over the extremities. Originally, they were not confluent but subsequently became so.

DR. ALEXANDER: Are there any suggestions as to the nature of these lesions?

DR. WOOD: The possibility of acute pancreatitis must be considered. I was told the lesions which this patient developed were comparable with the so-called Gray-Turner spots which are said to be pathognomic of acute pancreatitis.

DR. ALEXANDER: Dr. Scheff has already suggested that the patient had recurrent pancreatitis with acute hemorrhagic pancreatitis as a terminal episode; and if these skin lesions were indeed the type seen in acute pancreatitis, that diagnosis acquires further support.

In summary, it seems clear that we all agree that this patient had severe cardiovascular disease with aortic insufficiency possibly due to arteriosclerosis or to rheumatic involvement. In addition, it is thought that he had chronic pancreatitis with acute pancreatitis as a terminal episode.

Clinical Diagnosis: Aortic insufficiency due to arteriosclerotic and hypertensive cardiovascular disease or to rheumatic heart disease; chronic and acute pancreatitis.

PATHOLOGIC DISCUSSION

DR. MARGARET A. CARTER: There were petechiae and ecchymoses of the skin on the back and the thighs. The brain appeared normal externally except for an advanced degree of arteriosclerosis of the cerebral arteries. On section there was a focus of tan softening, about 2 cm. in diameter, in the right lentiform nucleus and anterior limb of the internal capsule. The lungs were heavier than usual, weighing 1,100 gm. The lower lobes were irregularly firm and moist and the margins showed focal emphysema. The heart weighed 985 gm. and was enlarged to about three times the normal weight for the

patient's size. The wall of the left ventricle was greatly thickened, and there were a few foci of fibrous thickening in the endocardium of both ventricles. The cusps of the four valves showed no significant pathologic lesions. The coronary arteries were thickened, and their intimal surfaces were irregular, flaky, and roughened. The lumens of the major branches were not significantly narrowed; however, in the smaller branches they were greatly decreased in diameter although not occluded. The aorta showed advanced stages of arteriosclerosis.

The kidneys were reduced in size; the capsules were stripped with slight difficulty to expose granular surfaces. On section the normal architectural pattern was obscured, the cortex thinned and the peripelvic fat increased. The major vessels of the kidney were firm and thickened but the orifices and lumens of the renal arteries were widely patent. The prostate showed a moderate amount of nodular hyperplasia. The bladder was thickened and its mucosa was congested. In the peritoneal cavity, confined to the right upper quadrant about the hepatic flexure of the colon, there were 15 cc. of a thin yellowish fluid. The adjacent peritoneal surfaces were congested, but showed no adhesions. Large numbers of petechiae and ecchymoses involved the mucosa of the entire colon and a few were present in the ileum and stomach. The liver was congested and enlarged to 1,970 gm. The large spleen weighed 400 gm. and its cut surface was soft and congested. The gallbladder was normal and the pancreas was not remarkable.

DR. ROBERT A. MOORE: It was apparent this man had extensive cardiovascular disease with an advanced degree of arteriosclerosis, particularly of the aorta, coronary and cerebral vessels, and the medium-sized and smaller renal vessels but not the main renal arteries.

The heart weighed 985 gm. and was hypertrophied and dilated. The fact that the heart had undergone a great degree of hypertrophy in the presence of advanced arteriosclerosis of the coronary arteries raises the question of the limiting factors in cardiac hypertrophy. As a general rule, the larger hearts show less arteriosclerosis, for otherwise there is insufficient nourishment for the increased amount of myocardium. In this instance sections of the arteries showed a definite narrowing of the lumens; yet there was three times as much heart muscle, first to be manufactured and second to be nourished by a decreasingly efficient coronary circulation.

There was also a focus of encephalomalacia in the right basal ganglia and the adjacent internal capsule that was of an arteriosclerotic etiology. This man, therefore, manifested lesions of arteriosclerotic cardiovascular disease in the heart, kidney and brain.

Figure 1 is of a section from the area of encephalomalacia; it demonstrates particularly well that the arteriolar disease had involved the cerebrum extensively. All the medium-sized and smaller vessels of the brain showed thickening, and in the one illustrated there was splitting apart of the wall, with edema in the resulting space. That, in other words, was the necrotic vascular change of malignant hypertension. Figure 2 shows the histologic characteristics of the encephalomalacia. There was softening, breaking apart and necrosis of the substance with the formation of gitter cells throughout the region. The histologic appearance of this lesion was compatible with a focus twelve days old, and its location anterior to the genu of the internal capsule correlated well with the history of a clearing hemiplegia; the adjacent corticospinal tracts were probably involved only by a reversible edematous reaction.

In the kidney (Fig. 3) the smaller arteries and arterioles showed a high grade of arterial and arteriolar sclerosis. The interstitial and glomerular changes, on the other hand, were minimal; there was only a small amount of fibrosis and destruction of renal parenchyma despite an anatomically markedly restricted blood supply. Figure 4 shows the cortex and capsule of the adrenal gland with the cells of the zona glomerulosa to the right. Here there were small vessels with hyalin walls that had undergone necrosis. This necrosis, however, had not reached the point that it had attracted leukocytes. It represented a very early stage of malignant hypertension. In Figure 5 an interesting phenomenon is shown, although I personally do not attach etiologic significance to the change depicted. It is a finding that is not uncommon in patients with long-standing hypertension. On the left is a section of the adrenal gland from this case; it includes a cross section of the central vein which shows tremendous hypertrophy of the muscular wall. In the right side of the illustration there is a normal central vein at exactly the same magnification that is three or four times thinner. This hypertrophy of

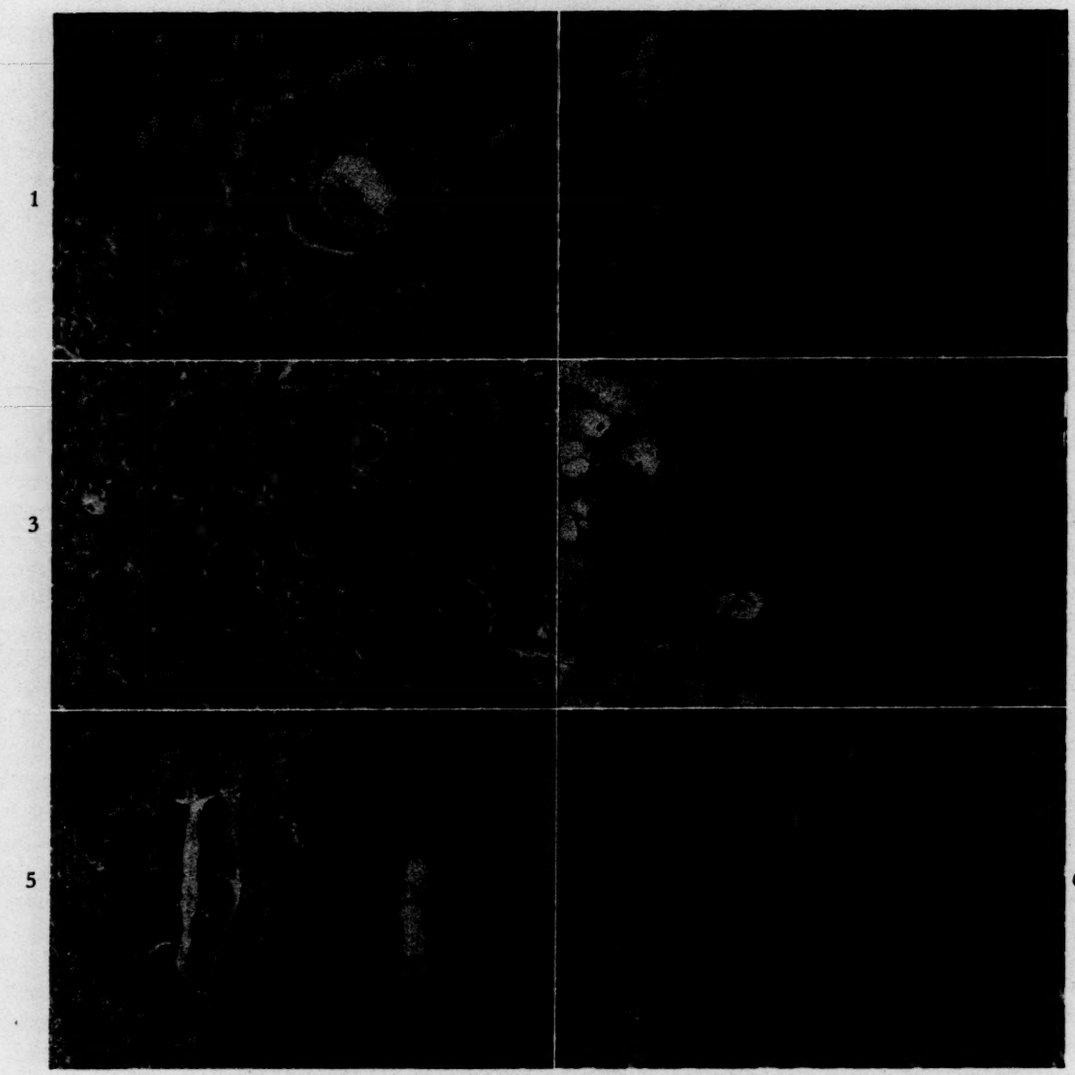

FIG. 1. Advanced arteriolosclerosis in the brain; the wall of this vessel in a focus of encephalomalacia was hyalinized and split as can be observed in malignant hypertension.
FIG. 2. Macrophages in a focus of encephalomalacia that was probably responsible for the episode of hemiplegia twelve days before the patient's death.
FIG. 3. The kidney with advanced thickening and sclerosis of all smaller arteries and arterioles but with few of the secondary nephrosclerotic changes in the interstitial tissue, glomeruli and tubules that usually follow arteriolosclerosis in the kidney.
FIG. 4. Advanced thickening, hyalinization and early necrosis of arterioles in the capsule of the adrenal gland.
FIG. 5. Left: the greatly thickened muscular wall of the central vein of the adrenal from this case; right: a normal central vein at the same magnification for comparison. Lesser degrees of this change are found not uncommonly in cases of hypertension.
FIG. 6. Acute bronchopneumonia, present particularly in the lower lobes.

the wall of the central vein of the adrenal has formed the basis of some theories concerning the relationship of the adrenal gland to hypertension.

Figure 6 is of a section of lung. This patient had foci of pneumonia throughout various parts of the lung. None of the areas was cultured, but bacterial stains revealed gram-positive cocci associated with this nodular pneumonia.

Concerning the matter of dilatation of the aortic ring, we have figures from some work done a number of years ago at the New York Hospital on the relationship of the cardiac rings to the

size of the heart. The normal heart weighs less than 350 gm., and the sizes of the tricuspid, pulmonary, mitral and aortic rings according to this study are less than 11.9 cm., 7.6 cm., 9.8 cm. and 7.4 cm., respectively. If the heart weighs 800 gm., which is as high as these figures go, the measurements are 14.2 cm., 8.7 cm., 10.5 cm. and 7.9 cm., respectively. The heart in this case weighed 985 gm. and the tricuspid valve was 13.5 cm., the pulmonary 9.0 cm., the mitral 11.0 cm. and the aortic 9.0 cm. It was also determined in the study to which I refer that in the presence of normal leaflets, despite a dilated ring, the valve was always adequate to close the orifice. This observation was made by measuring the ring, photographing the cusps at a known magnification, and from the photograph calculating by means of a planimeter the area of the valve leaflets. Of course, there remained the problem of the lack of elasticity of dead tissue. In this case it seems the aortic ring particularly was enlarged, but as the valve leaflets were normal we would have assumed they were sufficient to close the orifice of the aorta.

In regard to the patient's abdominal pain, there is no concrete anatomic explanation. There was an advanced degree of chronic passive congestion of the liver and spleen. Also, there were extensive petechiae and ecchymoses throughout the gastrointestinal mucosa which apparently accompanied sufficient injury to the wall of the colon that a slight bland peritonitis developed in the right upper quadrant, but there was no evidence that this process was of more than a few hours' duration. There was arteriosclerosis of the vessels in the pancreas with very slight necrosis and a few interstitial lymphocytes, but not enough to warrant the diagnosis of acute pancreatitis. The gallbladder was normal.

In summary, it seems this patient had had satisfactorily compensated hypertension for several years. The heart muscle responded well and developed a high degree of hypertrophy, but the episode of failure four years before death indicated that damaging lesions were developing and the limit of tolerance was being approached even then. About twelve days before his death a lesion developed in the brain due to the vascular disease, and then near the end of life his kidneys failed. One has the impression on inspecting the markedly sclerotic vessels of the kidneys that there should be a typical necrotic arteriole somewhere in the sections. I hesitate to go so far on just the evidence at hand, but it seems he was on the verge of developing malignant hypertension; if he had outlived the bronchopneumonia a few more days, he might have developed the full-blown picture.

DR. ALEXANDER: We certainly were wrong in our assumption that this patient had pancreatic disease. Could the skin manifestations have been due to uremia?

DR. WOOD: Certainly purpura develops in uremia but the lesions which this patient exhibited did not look like the purpura which I have seen in uremia. Similar lesions have been described in patients with myocardial infarction in shock, but Dr. Rouse has assured us that this patient's blood pressure was never at shock levels. Dr. Rouse, what did you think the skin lesions were due to?

DR. ROUSE: I thought they were the Gray-Turner spots of acute pancreatitis.

Final Anatomic Diagnoses: Arteriosclerosis of the aorta, coronary and cerebral arteries; encephalomalacia in the right basal ganglia; arteriolar nephrosclerosis; hypertrophy and dilatation of the heart; chronic passive congestion of the lungs, liver and spleen; bronchopneumonia.

Acknowledgment: Illustrations were made by the Department of Illustration, Washington University School of Medicine.

Research Society Abstracts

American Federation for Clinical Research

ABSTRACTS OF PAPERS PRESENTED AT THE EASTERN SECTIONAL MEETING IN WASHINGTON, D.C., DECEMBER 9, 1950

COMPARISON OF FAT TOLERANCE AS DETERMINED BY ALIMENTARY LIPEMIA IN NORMAL INDIVIDUALS AND IN PATIENTS RECOVERING FROM ACUTE MYOCARDIAL INFARCTION. *Leon Schwartz, M.D., Asher Woldow, M.D. (by invitation) and Richard A. Dunsmore, M.D. (by invitation), Philadelphia, Pa.*

When fasting subjects are given a standard fat meal, a measurable increase in serum turbidity (lipemia) as determined in a photoelectric colorimeter is noted in three to five hours after the test meal. In a preliminary study fifteen patients recovering from acute myocardial infarction showed fasting serum turbidity levels varying from 37 to 50 units (using distilled water blank) with an average value of 44 units. Three hours and five hours after a standard fat meal serum turbidity had risen to levels varying from 90 to 375 units and 56 to 250 units, respectively, and averaged 207 and 146 units, respectively. A control group of ten apparently normal subjects, four of whom were fifty years of age or older, showed fasting turbidity levels which varied from 26 to 40 units and averaged 30 units. Three hours and five hours after the test meal turbidity levels ranged from 38 to 79 units and 33 to 73 units, respectively, and averaged 59 and 53 units, respectively.

These preliminary results suggest that the study of serum turbidity changes which follow a standard fat meal may be useful in evaluating the possible role of impaired fat tolerance in the etiology of atherosclerosis.

CLINICAL COMPARISON OF INTERMEDIATE INSULINS IN THE CONTROL OF SEVERE DIABETES. *Samuel J. N. Sugar, M.D. and Louis K. Alpert, M.D., Washington, D.C.*

Eight patients with severe diabetes, requiring 30 or more units of insulin a day, were given equivalent doses of three types of insulin: globin zinc insulin, 2:1 mixtures of regular and protamine zinc insulin, and NPH 50. Each type of insulin was used for periods of four to seven days. Diet and physical activity were maintained at a constant value. Blood sugars were determined before meals at 8 A.M., at noon, and at 4 P.M. Fasting blood sugar levels of 150 mg. per cent or less occurred about one and one-half times more often with globin insulin and 2:1 mixtures as with NPH50. At noon blood sugar values below 200 mg. per cent were observed with equal frequency with all three types of insulin. At 4 P.M. the intensity of action was approximately equal with all the types of insulin used. Hypoglycemic reactions occurred only on two occasions, both times with 2:1 mixtures. The most satisfactory control, without hypoglycemic reactions, was obtained with the use of globin zinc insulin.

NPH50 insulin gave good control but fasting blood sugar values below 150 mg. per cent were obtained only two-thirds as often as with either globin zinc insulin or 2:1 mixtures.

Although 2:1 mixtures gave the best control on the basis of blood sugar levels, the occurrence of hypoglycemia in two instances would place this type of insulin in a less desirable position in relation to the other two which were used.

NEW METHOD FOR THE DIAGNOSIS OF HYPERTHYROIDISM USING RADIOACTIVE IODINE. *Solomon Silver, M.D. and Mack H. Fieber, M.D., New York, N. Y.*

Previous methods employing I^{131} in the diagnosis of hyperthyroidism have depended upon the increased avidity of the gland for inorganic iodide. This phenomenon, however, is only incidental to the disturbance from which the patient suffers, namely, the increased secretion of thyroid hormone into the circulation. The two events although related need not be parallel.

The blood levels of I^{131} have been determined after tracer doses for a period of ninety-six hours. It has been clearly shown that after forty-eight hours the level of circulating I^{131} is markedly increased in hyperthyroidism as compared with normals and that this increase is due to protein-bound I^{131}. The range of values in over 100 cases is such that there is no overlap between the

highest values in the normals and the *lowest* values in the hyperthyroids. This laboratory procedure appears to offer the best method available for the diagnosis of hyperthyroidism.

RENAL CLEARANCES OF PENICILLIN, PHENOLSULPHONPHTHALEIN (PSP) AND PARA-AMINOHIPPURATE (PAH) MODIFIED BY "BENEMID." *William P. Boger, M.D., Walter V. Matteucci, M.D. (by invitation) and Nelson H. Schimmel, M.D. (by invitation), Philadelphia, Pa.*

It has been established that penicillin, phenolsulphonpthalein (PSP) and paraaminohippurate (PAH) are rapidly eliminated. The rate these substances are cleared measures renal plasma flow. It has been shown by others that PAH and diodrast competitively inhibit the excretion of penicillin by the renal tubules. The desirability of inhibiting the renal tubular secretion of penicillin is obvious but until recently this has been impractical. A new compound, para (di-n-propylsulfamyl)-benzoic acid, "benemid" in small daily doses (2 gm.) has been shown to produce reversible inhibition of the tubular excretion of penicillin. It was found that benemid (1) reduces the normal renal clearance of penicillin within fifteen minutes to glomerular filtration rate as measured by creatinine clearance, (2) reduces the renal clearance of PSP to one-fifth the normal clearance and (3) reduces the clearance of PAH, but not to glomerular filtration rate. The reversibility of the tubular inhibition produced by "benemid" is demonstrated by use of the fifteen-minute PSP excretion test. The failure to depress renal tubular secretion of PAH as strikingly as is the case with penicillin and PSP is interpreted as representing greater affinity of PAH for the enzyme transport mechanism whereby these substances are secreted.

USE OF CATION EXCHANGE RESINS IN THE CONTROL OF SODIUM BALANCE. *Richard H. Lyons, M.D., John Horn, M.D., Don C. Nouse, M.D. and John McCabe, M.D., Syracuse, N. Y.*

This study was initiated to evaluate the effectiveness and safety of the use of cation exchange resins in control of sodium balance. The resin used was a mixture of carboxylic and potassium amberlite resin. This was given in dosages of 15 gm. three or four times daily. Patients were maintained on 2,500 cc. fluid intake daily and a diet estimated at 40 per cent greater than basal caloric estimation. Daily basal weights were recorded on a special scale of 100 kg. capacity and of an accuracy of 10 gm. Serum and urinary potassium and sodium determinations were done at daily intervals. The resins were given to seven subjects with edema; definite control of sodium absorption was accomplished. There were no observed ill effects. Four normal subjects were observed with varying sodium intake and 60 gm. of mixed resins were effective in control of sodium intake up to 121 mEq. daily. Three subjects have been followed by therapy with resins over an interval of four to five months. No laboratory or clinical evidence has been observed to contraindicate the use of mixed resins in control of body sodium balance. Resins have been shown effectively to control sodium absorption from the gastrointestinal tract.

ELECTROLYTE AND WATER CONTENT OF CARDIAC AND SKELETAL MUSCLE IN NORMALS, VENTRICULAR HYPERTROPHY AND INFARCTION. *L. C. Alexander, M.D. (by invitation), A. J. Boyle, M.D. (by invitation), L. T. Iseri, M.D., R. S. McCaughey, M.D. (by invitation) and G. B. Myers, M.D., Detroit, Mich.*

In fifty-three autopsies muscle blocks free of gross fat were obtained from the center of the interventricular septum from three standard left ventricular sites and from the pectoralis major. All specimens were analyzed for sodium and potassium and for total water. In approximately two-thirds of the cases analyses were also made for phosphorus, magnesium, calcium and copper, and chemical analysis was made for chloride. The cases were classified from the standpoint of cardiac findings at autopsy into the following groups: normal, nineteen cases; recent infarction, eight and left ventricular hypertrophy, twenty-six. The latter group was subdivided into fourteen digitalized and twelve non-digitalized cases.

The average values for normal myocardium per wet tissue were: H_2O, 78.7 per cent; Cl, 0.153 per cent; Na, 0.117 per cent; K, 0.322 per cent; Mg, 0.021 per cent; P, 0.212 per cent; Ca, 0.007 per cent; and Cu, 0.0003 per cent. Skeletal muscle was comparable in calcium, magnesium and phosphorus content, slightly lower in water and sodium, distinctly lower in chloride and copper, but significantly higher in potassium.

The average values in uncomplicated left ventricular hypertrophy were almost identical with the normal. There was no significant difference in the hypertrophied but digitalized hearts except in respect to potassium. The average potassium content in six digitalized but com-

pensated hearts was 0.281 per cent, and in eight digitalized but decompensated hearts it was 0.272 per cent. The potassium content of skeletal muscle was correspondingly reduced in the digitalized and decompensated cases.

Recently infarcted myocardium was slightly lower in water content, significantly lower in potassium (0.189 per cent), magnesium (0.011 per cent) and phosphorus (<0.100 per cent) but slightly higher in calcium and copper, and significantly higher in sodium (0.195 per cent) and chloride (0.260 per cent).

STUDY OF SODIUM AND WATER METABOLISM IN PATIENTS WITH AND WITHOUT LIVER DISEASE DURING CONTINUED ADMINISTRATION OF PITRESSIN TANNATE. *Richard H. Lyons, Jay F. Harris and Charles W. Lloyd, Syracuse, N. Y.*

Seven patients were studied for comparative responses to daily pitressin administration over a short period of time in an attempt to distinguish the various effects of antidiuretic hormone upon salt and water balance. Of those studied two were considered as normal, three had Laennec's cirrhosis, one infectious hepatitis and one alcoholic. Intake of fluid and sodium were controlled. Twenty-four-hour urine samples were collected daily for sodium content. All patients were begun on pitressin tannate in oil, 5 u intramuscularly daily, and their responses compared.

The three cirrhotics evidenced definite increased response to pitressin as compared with the two controls by gaining double the weight from fluid retention by the sixth day. The cirrhotics also evidenced a remarkable drop in the plasma sodium level while the controls showed only minimal effect. The dual effects of pitressin, i.e., antidiuresis and naturesis, were illustrated in various degrees and usually explained the development of hyponatremia. It is significant that frank and subclinical edema could be produced while at the same time producing a falling plasma sodium level.

The patient with infectious hepatitis followed the pattern of the controls while the alcoholic showed responses similar to the cirrhotic.

REACTION OF HUMAN SERUM WITH A QUATERNARY AMMONIUM SALT: RESULTS OF A SERIAL STUDY IN ACUTE RHEUMATIC FEVER. *R. F. Jacox, M.D. (introduced by L. E. Young, M.D.), Rochester, N. Y.*

Under controlled conditions of pH, salt concentration and serum to quaternary ammonium salt (QAS) ratio, human serum develops turbidity after addition of QAS. In normal sera a narrow range of variation is found whereas marked increases of turbidity develop in sera of individuals with acute rheumatic fever, pneumonia, carcinoma or other unrelated pathologic disorders. The reacting component in serum with QAS is neither albumin, mucoprotein nor gamma globulin but is probably a component of fraction IV-1.

Observations on a group of twenty patients with acute rheumatic fever reveal that the serum-QAS reaction is an extremely sensitive measure of rheumatic activity. Correlative studies in this group were made with respect to erythrocyte sedimentation rate, bactericidal activity of serum for B. subtilis and serum-QAS turbidity. The latter technic was not only the easiest of these assays to determine but reflected most accurately the degree of rheumatic activity.

TURBIDIMETRIC ESTIMATION OF GAMMA GLOBULIN IN RHEUMATIC DISEASES. *W. R. Merchant, M.D., H. J. Zimmerman, M.D. and T. McP. Brown, M.D., Washington, D.C.*

Abnormal serum proteins, depression of the albumin and an increase in the globulin fraction, have been frequently noted in rheumatoid arthritis. The increase in the euglobulin fraction, chemically determined, seems correlated with elevation of the gamma globulin level as determined electrophoretically. The apparent correlation of these findings with clinical disease activity suggested that the determination of the serum protein fractions might serve as a diagnostic adjunct as well as a possible index of the response to therapy. The recent introduction of a simple turbidimetric estimate of the gamma globulin level by Kunkel provided a practical method for obtaining this information.

Approximately 500 determinations were carried out on eighty-three patients with various arthritic diseases. The average values were higher in forty-two patients with rheumatoid arthritis as compared to other arthritic states and normal controls. Sixteen patients with spondylitis, with an occasional exception, had lower values. Three patients with Reiter's disease had both normal and abnormal values. Thirteen patients with gout and hypertrophic arthritis had normal values.

In rheumatoid arthritis abnormal values could usually be correlated with disease activity. However, when the zinc sulfate turbidity level is normal in active disease, the albumin and globulin levels are essentially normal. No patient with

active disease had both a normal zinc sulfate turbidity and sedimentation rate. With interesting exceptions, elevated sedimentation rates paralleled abnormal zinc turbidity values. There was no constant quantitative relationship between gamma globulin values obtained by electrophoresis and those with the turbidimetric technic.

QUANTITATIVE STUDY OF SERUM ANTIPROTEOLYTIC ACTIVITY IN DISEASE WITH SEPARATION OF TWO SERUM PROTEOLYTIC INHIBITORS. *N. R. Shulman and H. J. Tagnon, New York, N. Y.*

Increased serum antiproteolytic activity has been observed in various diseases. Published reports make no distinction in serum inhibition of the enzymes, trypsin, chymotrypsin and plasmin. The apparent assumption is that a single inhibitor for the three enzymes occasionally increases in disease.

A method of measuring proteolytic activity using a radioactively tagged substrate was developed in this laboratory to permit highly sensitive measurements in media such as serum. Normal serum inhibition of trypsin, chymotrypsin and plasmin was determined and compared with that of patients with carcinoma and patients with tuberculosis. Many patients showed abnormal inhibition. Those abnormal sera showing increased trypsin inhibition also showed increased chymotrypsin inhibition. Increases in inhibition were quantitatively equal for these two enzymes, and the magnitude of increase was similar in carcinoma and tuberculosis.

Sera with increased trypsin and chymotrypsin inhibition showed no increased plasmin inhibition. This new finding was evidence for at least two separate inhibitors. It was corroborated by ammonium sulfate fractionation of normal and abnormal sera. Plasmin inhibition was greatest in the globulin fraction whereas inhibition of trypsin and chymotrypsin was greatest in the albumin fraction. Fractions of normal and abnormal sera contained the same relative proportion of inhibition for the three enzymes.

STREPTOCOCCIC ANTIHYALURONIDASE IN ACUTE RHEUMATIC FEVER, ACUTE RHEUMATOID ARTHRITIS AND ACUTE GLOMERULONEPHRITIS; A MODIFIED TEST. *Robert T. Thompson, M.D. and Florence M. Carpenter, B.A. (by invitation), Cincinnati, O.*

The value of serum streptococcic antihyaluronidase titers for the diagnosis of acute rheumatic fever has been demonstrated by Quinn and Harris and Harris using the mucoprotein clot prevention test, with two-fold serial dilution of patient's serum. Diluents used by these authors were distilled water and physiologic saline, respectively. These diluents cause a partial loss in the potency of hyaluronidase in high dilution. Therefore, the present study was carried out using a modified mucoprotein clot prevention test with 1.0 per cent proteose peptone in physiologic saline as the diluent, and the patients' serum was titrated by the more accurate method of measured dilution. Group A streptococcic antihyaluronidase serum titers of 417 or greater were found in thirty-six of forty-two patients with acute rheumatic fever, in eleven of fifteen patients with acute glomerulonephritis, in one of twenty patients with acute rheumatoid arthritis and in three of fifty "normal" subjects. Serum titers of 900 or greater were found in twenty-nine of these rheumatic fever patients and in nine of these glomerulonephritis patients. Serum titers of 278 or less were found in seven patients with inactive rheumatic heart disease, four patients with acute benign idiopathic pericarditis and three patients with acute gonococcic arthritis.

QUANTITATIVE EVALUATION OF THE INCREASE IN BLOOD FLOW IN THE HUMAN FOOT AFTER HEXAMETHONIUM AS COMPARED TO PROCAINE BLOCK. *Harold W. Schnaper, M.D., Robert L. Johnson, M.D., Edward Tuohy, M.D. and Edward D. Freis, M.D., Washington, D.C.*

Clinical and experimental studies in this laboratory indicated that the new ganglionic blocking agent hexamethonium (C_6) produced greater vasodilatation in the feet than any previously known compound.

Using the Abramson venous occlusion plethysmograph, foot blood flow was determined in normal volunteers both after C_6 and again after lumbar epidural block and/or spinal anesthesia to the level of the iliac crests. Environmental temperatures were maintained at a constant level of 68 to 69°F. The effectiveness of block was corroborated by skin temperature changes and digital plethysmography.

In all instances except one the increase in foot blood flow approached or exceeded the increase following epidural or spinal block. These studies indicate that C_6 is not only a more potent peripheral vasodilator than any other previously studied compound but also produces a vasodilatation of the feet comparable to regional block (complete sympathetic blockade).

EXPERIENCE WITH THE NEW ANTICOAGULANT, PARITOL-C. A PRELIMINARY REPORT. *Benjamin Manchester, M.D., Elizabeth H. Hill, M.D., Boris Rabkin, M.D. and Clifford B. Sagemen, M.D., Washington, D.C.*

During the past year Paritol-C, a new synthetic anticoagulant, has been employed for its rapid and sustained effect in prolonging the blood clotting time. Paritol-C is a polysulfated ester of dihydromanuronic acid derived from kelp seaweed. Its action is similar, in some respects superior, to heparin.

Thirty-seven subjects received Paritol-C for twenty-four to seventy-two hours; one individual received the drug for seven days. The dosage was calculated according to weight; 5 mg. per kg. of body weight. It was given intravenously through a No. 25 or No. 26 gauge needle at intervals of eight to twelve hours. Clotting time was obtained before the administration, thirty minutes, one, two, four, six and eight hours after the intravenous injection of Paritol-C. The Lee-White two-tube clotting time was used. Effective anticoagulation was considered when the clotting time was two to three times the control value (eighteen to thirty minutes). In all instances dicumarol was started simultaneously with intravenous Paritol-C in the usual doses.

The results indicate that this drug is a rapidly acting anticoagulant with prolonged action and without the rebound phenomenon of heparin. It prolongs the clotting time within thirty minutes to three to four times the control and maintains an active antithrombotic level for six to eight hours in 75 per cent of patients. It offers promise of gaining a place beside heparin as a rapidly acting agent in prolonging clotting time.

CLINICAL EVALUATION OF THE BALLISTOCARDIOGRAM. I. PRELIMINARY REPORT. *Marvin Moser, M.D. (by invitation), Robert C. Taymor, M.D., Leon Pordy, M.D., Kenneth Chesky, M.D. and Arthur M. Master, M.D., New York, N. Y.*

Over 300 cases have thus far been studied by means of the simplified displacement type ballistocardiogram which employs a photoelectric cell, as suggested by Dock. The first 175 cases being reported include: (1) normal persons, (2) patients with negative resting electrocardiograms but with positive Master "Two-step" exercise electrocardiograms, (3) patients with hypertension and (4) patients with previous myocardial infarction.

The fifty normal persons varied in age from eighteen to sixty-two years. The ballistocardiographic pattern was normal in forty-five, abnormal in four and borderline in one. A study of the ballistocardiogram in forty-five patients with normal resting electrocardiograms but positive exercise electrocardiograms showed abnormal resting ballistocardiograms in thirty-four, normal in ten and borderline in one. Seven of these patients with normal resting ballistocardiograms had records taken after exercise and the ballistocardiogram became abnormal in five. Of forty cases of essential hypertension and hypertensive heart disease with normal or abnormal electrocardiograms at rest only one was found to be normal. Of forty cases with previous myocardial infarction three had normal ballistocardiograms at rest.

An excellent correlation has thus far been obtained between the ballistocardiographic and electrocardiographic patterns. It is believed that this simplified type of ballistocardiograph may be employed clinically and will probably prove to be of value in the routine evaluation of cardiac patients.

DUODENAL "STUMP ULCER": ITS DESCRIPTION AND DIAGNOSIS. *Stanley H. Lorber, M.D. and Harry Shay, M.D. (by invitation), Philadelphia, Pa.*

The presence of an active ulcer in the duodenal stump has not been previously emphasized among the complications of gastric resection even though perforation of the duodenal stump continues to be an infrequent but well recognized hazard of such surgery. Subsequent examination of patients subjected to partial gastrectomy has revealed a small percentage who continue to have ulcer symptoms and in whom no evidence of gastric, marginal or jejunal ulcer can be demonstrated. Two patients recently observed experienced a shift in location of their ulcer pain from the epigastrium to the right upper quadrant following subtotal gastric resection. Repeated routine roentgen examinations of the upper gastrointestinal tract failed to reveal the cause of their pain. Because of the location of the pain and persistence of hyperacidity in both patients, ulceration in the afferent (duodenal) loop was suspected. Utilizing a special intubation technic the afferent loop was adequately filled with barium. In each case fluoroscopy and roentgenograms demonstrated a "stump ulcer" which coincided with the area of pain.

EFFECT OF THE CITROVORUM FACTOR IN PERNICIOUS ANEMIA. *R. R. Ellison (by invitation), S. Wolfe (by invitation), H. Lichtman, V. Ginsberg and J. Watson, Brooklyn, N. Y.*

The effect of the citrovorum factor (C. F.) administered parenterally to six patients with Addisonian pernicious anemia has been studied. The C. F. used was material containing 20 million units (3 mg.) per cc. Two patients given 5 million units daily had about half-optimal reticulocyte and red cell rises. Of two patients given 10 million units daily one had a maximal red cell and reticulocyte response and the other suboptimal. Clinical improvement was quite satisfactory. It is thought that 10 million units (1.5 mg.) may be approximately equivalent to 1 U.S.P. unit of liver extract but further study is indicated.

Two patients were given aminopterin and PGA parenterally for ten days. In one patient there was no reticulocyte rise until C. F. was substituted for the PGA in a second ten-day period. In the other patient (on half the dose of aminopterin and the same amount of PGA) there was a moderate reticulocyte rise in the first ten-day period but a secondary reticulocyte peak occurred in the second ten-day period in which C. F. was substituted for PGA. It is thought therefore that C. F. is more effective than PGA in reversing the inhibitory effect of aminopterin. One patient who was given 10 million units of C. F. intrasternally showed no enhanced local maturing of megaloblasts at the site of instillation.

IRON METABOLISM IN HEMOCHROMATOSIS. II. INTRAVENOUS IRON TOLERANCE CURVES. *Stanley Edward Gitlow, M.D. and Milton R. Beyers, M.D., Brooklyn, N. Y.*

A high serum iron with saturation of the iron-binding capacity has been suggested as being diagnostic in hemochromatosis. Intravenous iron tolerance curves were performed in normals, patients with various hematologic disorders, those with cirrhosis and diabetes without hemochromatosis and in four patients with hemochromatosis. The peak of the iron tolerance curve reflects the serum iron-binding capacity.

Three of the four patients with hemochromatosis had high fasting serum iron with a high degree of saturation of the iron-binding capacity. One had a normal fasting level with saturation of the iron-binding capacity only slightly above normal. In two of the four, including the latter, the serum iron fell more rapidly from the peak values than normally and reached subfasting levels, a condition not found in normals. This reflects an increased avidity of the tissues for serum iron, suggesting a further defect in hemochromatosis besides that of the increased gastrointestinal absorption of iron. Determinations of fasting serum iron and serum iron-binding capacity are apparently not always sufficient for the diagnosis of this disease.

Pernicious anemia in relapse and polycythemia vera may reveal similar abnormalities in the iron tolerance curve.

STUDIES ON NITROGEN AND PHOSPHORUS METABOLISM WITH RELATION TO THE HEMATOLOGIC RESPONSE IN PERNICIOUS ANEMIA PATIENTS AFTER CRYSTALLINE VITAMIN B_{12}. *G. Watson James, III, M.D. and Lynn D. Abbott, Jr., Ph.D. (by invitation), Richmond, Va.*

In view of the response in hemoglobin synthesis caused by crystalline vitamin B_{12} in patients with pernicious anemia it was considered desirable to correlate hematologic observations with nitrogen and phosphorus balance studies before and during clinical remission in an attempt to elucidate further the metabolic effects of this substance. Four previously untreated patients with pernicious anemia were studied for several weeks on a constant diet. Positive nitrogen balance as great as 6 gm. per day was produced by vitamin B_{12} in each case. When negative nitrogen balance existed during the control period, it promptly became positive. Individual patients made from 18 to 37 gm. of hemoglobin per day for an average period of eleven days. Nitrogen required for hemoglobin formation alone equalled or exceeded the total nitrogen retained during the corresponding period.

The following sequence of biochemical changes was observed: First, a prompt and striking decrease in urinary phosphorus which preceded any change in reticulocyte count; second, increased excretion of uric acid during the upswing of reticulocytes; and third, increased excretion of phosphorus during the period of greatest reticulocytosis.

These clinical observations suggest that among the first metabolic effects of vitamin B_{12} is a profound influence on nucleoprotein synthesis.

WEST NILE AND ILHEUS VIRUS INFECTIONS IN PATIENTS WITH NEOPLASTIC DISEASE. *Chester M. Southam, M.D. and Alice E. Moore, M.D., New York, N. Y.*

West Nile virus and Ilheus virus have been shown by Koprowski and Norton to have an

oncolytic effect against certain mouse tumors. West Nile has been inoculated intramuscularly or intravenously into twelve patients with anaplastic carcinoma, ovarian carcinoma or mammary carcinoma. Viremia persisting forty-eight hours or longer occurred in four patients but did not persist more than one week. Virus has not been detected in urine, stool or throat washings. In one patient virus was present in the carcinoma forty-eight hours after it had disappeared from the blood. In two patients there was slight fever (38°C.) between the third and tenth day and a higher febrile spike (40°C.) during the third week. One patient had no temperature elevation. The fourth died of uremia fifteen days after inoculation. There were no subjective complaints or neurologic signs attributable to the virus.

Ilheus virus has been inoculated in the same manner into seven patients of whom four have had persistent viremia. Diagnoses were mammary carcinoma, acute monocytic leukemia, subacute lymphatic leukemia and osteogenic sarcoma. One of the patients died on the sixteenth day after inoculation and had had no signs or symptoms attributable to virus by that time. The other three patients had fever for two to five days (39° to 40°C.) during the third week. During this febrile period all complained of vague generalized discomfort and exhibited drowsiness alternating with apprehension which suggested a mild encephalitis. The only definite objective neurologic change was loss in one patient of all deep tendon reflexes during the febrile period, with rapid recovery during the next three days.

The patient with lymphatic leukemia had disappearance of hepatosplenomegaly and adenopathy, marked subjective improvement and weight gain, but no improvement in blood or marrow. All other patients showed no improvement.

THE COMMON COLD: CLINICAL STUDIES WITH FOUR AGENTS PRODUCING RESPIRATORY DISEASE ENTITIES IN HUMAN VOLUNTEERS. *Leon T. Atlas, M.D., Bethesda, Md.*

Four respiratory disease entities were characterized in human volunteers by the instillation of nasal washings from donors. MR-1, MR-2 and MR-3 viruses were cultivated in embryonated eggs; each reproduced in volunteers the disease suffered by the donor and studied in volunteers receiving the donor's washings. Transmission of MR-1 and MR-3 disease from inoculated to uninoculated contacts was demonstrated. A statistical evaluation of clinical findings for each disease will be presented.

Cultivated MR-1 virus produced disease in 168 volunteers. This illness was characterized by a twenty-four to seventy-two hours' incubation time, pharyngitis, laryngitis and post-nasal discharge. Two hundred volunteers receiving control fluids developed no illness. Twenty-four volunteers reinfected two and four weeks later with MR-1 developed colds as originally. MR-2 disease appeared twenty-four to ninety-six hours post-inoculation with rhinitis, headache and coughing. No resistance to MR-2 reinfection was demonstrable. MR-3 nasal washings produced characteristic illnesses in seventy-four of ninety-nine volunteers. This illness characteristically had a seventy-two to ninety-six hours' incubation, "running nose," cough and chest pains. Ninety-nine volunteer controls developed no disease. Resistance to MR-3 reinfection persisted over three months. MR-4 nasal washings produced disease in twenty-seven of fifty-eight men; a forty-eight to ninety-six hours' incubation, rhinitis, sneezing and resistance to reinfection over one month characterized MR-4 disease.

HERPANGINA—ETIOLOGIC STUDIES OF A SPECIFIC INFECTIOUS DISEASE. *R. J. Huebner, M.D., R. M. Cole, M.D., E. A. Beeman, M.D., J. A. Bell, M.D. and J. H. Peers, M.D., Bethesda, Md.*

A disease having the clinical and epidemiologic attributes of "herpangina" was found to be prevalent in the metropolitan area of Washington, D.C., in the summer of 1950. Etiologic studies of thirty-seven cases resulted in the isolation of viruses similar to the A group of Coxsackie viruses from thirty-two of the thirty-seven cases. Virus was demonstrated in the feces of thirty-one cases, in throat swabs of twenty cases and in anal swabs of twelve cases. Neutralizing antibodies against homologous virus developed in high titer in the serum of four cases that were tested. The viruses were classified in at least four immunologic groups and designated as H1, H2, H3 and H4. The viruses in these groups were found in nearly all frank cases of herpangina and in many persons having contact with frank cases of this disease. They were rarely found in persons not known to have recent contact with frank cases and rarely found in cases of other illnesses. It is hypothesized that these H viruses are the cause of herpangina.

EXPERIMENTAL POLYRADICULITIS IN MONKEYS. *Orville T. Bailey, M.D., Alwin M. Pappenheimer, M.D., F. Sargent Cheever, M.D. and Joan B. Daniels, B.S., Boston, Mass.*

In a routine attempt to demonstrate the virus of poliomyelitis in a child suspected of having the disease, fecal material was injected into Macaca mulatta monkeys. The lesions produced bore little resemblance to those of classical anterior poliomyelitis but presented a close analogy to the lesions of human polyradiculitis (Guillain-Barré). The changes in the spinal nerve roots and peripheral nerves consisted of neuronal degeneration with or without lymphocytic and mononuclear cell reaction. Further passage carried out by the injection of cord suspension was followed by the production of similar lesions in two of the six animals injected. The lesions in the monkeys were thus transmissible but it has been impossible to investigate by serologic means the relationship between the patient's illness and the lesions observed in the inoculated monkeys. Spontaneous occurrence of the lesions would seem to be excluded by their appearance in the course of inoculations years apart in monkeys obtained from different sources, by failure to find similar changes in a large number of monkeys used in our laboratory for other experiments and by the fact that their occurrence has not been described by others. The results of this study suggest the value of further investigation by virus technics in the search for the etiology of certain cases of human polyradiculitis.

USE OF RABBIT EYE AS TISSUE TO STUDY TUBERCULOSIS. *Paul A. Bunn, M.D. and Charles Adair, M.D., Syracuse, N. Y.*

Rabbit eyes have been used to study tuberculosis produced by tubercle bacilli H37Rv. Ocular disease in rabbits has many of the characteristics of human-type tuberculosis. Two major types of ocular disease have been developed, advanced, permanently destructive and minimal with healing. Size of infecting dose of tubercle bacilli decides the type; previous immunization, species of animal and method of inoculation have little influence. With the administration of a large inoculum advanced disease is produced in 73 per cent of instances. Small numbers of organisms result in reversible disease in 67 per cent of animals. In both types the course of the disease is self-limited and rarely extends beyond the orbit. The main purposes of the study were to develop accessible lesions that could be observed in life and to investigate the possibilities of using the lesion as a screen for tuberculostatic agents.

Streptomycin is capable of partially preventing ocular lesions if administered twenty-four hours prior to inoculation of organisms into the eye. When treatment with streptomycin is instituted from twenty-four to ninety-six hours after inoculation, it fails to alter appreciably the course of the tuberculous lesion although some effect in slowing the disease and reducing its acuteness is noted. With treatment delayed five or more days, when ocular tuberculosis is well established, no agent affects the disease significantly. No single other tuberculostatic material is as powerful as streptomycin in these observations. Seven other materials have been investigated and no one nor any combinations of these with streptomycin enhances the antimicrobial activity of streptomycin alone.

Case Reports

Pheochromocytoma with False Negative Benzodioxane Tests*

ROBERT E. MASON, M.D.
Baltimore, Maryland

THIRTEEN years ago when Howard and Barker wrote their excellent paper[1] on paroxysmal hypertension associated with pheochromocytomas, they were able to find only nineteen cases which were verified well enough for them to be certain as to the diagnosis. Since then reports of paroxysmal and sustained hypertension due to pheochromocytomas have appeared with increasing frequency and the total of well documented cases now exceeds one hundred. In addition to clear descriptions of the easily recognizable clinical picture tests have been described[2–5] which are capable of adding important confirmatory information and of establishing the diagnosis when the historical information is equivocal. Cahill and Aranow[6] have recently summarized much of the present information on this subject. There is no need at this time for further review of the subject as a whole, but individual cases should still be reported when they contribute important information. The following cases present data of interest particularly regarding some of the diagnostic tests:

CASE SUMMARIES

CASE I. P. M., a thirty-three year old white male clerk, presented himself in October, 1949, for investigation of "attacks of forceful heartbeat." His parents and six siblings were living; the father was diabetic. He had had an appendectomy in 1934 and a tonsillectomy in 1939. There had been no serious injury or infectious disease. He had enjoyed good general health all his life except for this present illness.

While on active duty with the Armed Forces in New Guinea in 1944 he experienced his first attack of forceful pounding heart action with an intense pounding headache. This attack lasted for only a few minutes, leaving him feeling quite normal at its termination. Similar attacks recurred at irregular intervals, averaging perhaps a week between attacks. Each attack began by his feeling that "something was going to happen" to him, followed by an increase in the force of his heart beat over an interval of ten to twenty beats. Somewhere in this period of increasing forcefulness a pounding vertical headache would begin and become almost intolerable at the height of the attack. In the early attacks he became quite frightened and noted some tachycardia but as he became more familiar with the attacks he found that there was not necessarily any change in the rate of the heartbeat, merely an increase in its force. He noted that if he were in bed during an attack the force of each beat would visibly shake the bed. He had been told that his face blanched during an attack. These attacks would last from one to thirty minutes and would die away gradually. He was unable to correlate them with mental or physical activity of any kind. During one attack in New Guinea his systolic blood pressure was recorded at 220. Because of the attacks he was returned to this country and finally discharged from the Armed Forces. The attacks continued with gradually increasing frequency until at the time he presented himself for examination he was averaging three a day. Despite these attacks he continued to carry out the duties of a responsible job.

Physical examination revealed the following: Temperature 99.2°F., pulse 80, blood pressure 164/90; height 73 inches, weight 186½ pounds. He was a tall, athletic, well nourished, intelligent white male who was not apparently ill or tense. The head was normal in all respects; the thyroid was of normal size and consistency; lungs and heart were not remarkable. No organ, mass or tenderness was palpated in the abdomen. The

* From the Department of Medicine, Johns Hopkins University School of Medicine, Baltimore, Md.

McBurney scar was well healed. The genitalia, rectum, extremities and central nervous system were normal to examination.

Laboratory data were as follows: *Blood:* corrected sedimentation rate, 10 mm.; hemoglobin 14.6 gm.; hematocrit 46.2 per cent; white blood count 8,800; polymorphonuclears 58; juvenile forms 1; eosinophils 5; basophiles 1; lymphocytes 25; monocytes 10. Cholesterol was 314 mg. per cent and non-protein nitrogen 34 mg. per cent. The basal metabolic rate was plus 17. *Urinalysis:* specific gravity 1.024; no sugar, albumin, red blood cells or casts and only rare white blood cells. The glucose tolerance test with the oral administration of 100 gm. of glucose revealed the following in milligrams per cent: fasting, 130; 30 minutes, 146; 1 hour, 120; 2 hours, 88; 3 hours, 108; 4 hours, 96. An electrocardiogram, x-rays of the chest and abdomen, and intravenous pyelograms demonstrated no abnormalities. A laminogram 8 cm. from the back revealed a round mass at the superior pole of the right kidney approximately 4 cm. in diameter.

On November 7, 1949, after four days of observation and testing, Drs. Hugh J. Jewett and Mark M. Ravitch explored the adrenal area through a transverse upper abdominal incision. In the right adrenal a tumor was found. Manipulation of this tumor produced an immediate rise in blood pressure. Thereafter great care was taken to avoid handling the tumor until its blood supply was ligated. Some normal adrenal tissue was left intact at the medial pole. There was no evidence of metastasis from the tumor which appeared grossly to be completely encapsulated. The left adrenal and the course of the aorta were explored and no further tumor was found. The operative and postoperative blood pressure recordings were remarkably uniform. During the operation the pressure varied from 140/70 at the onset to 190/118 during manipulation of the tumor, never falling below 130/74. The lowest blood pressure on the first postoperative day was 120/58. By the day of discharge this had settled further and was maintained at 110/60. As prophylactic preoperative preparation the patient received adrenal cortical extract 5 cc. intramuscularly the morning of the operation. It was unnecessary to give any adrenalin during or following operation to support the blood pressure, nor was there any evidence of adrenal cortical insufficiency after operation, clinically or chemically.

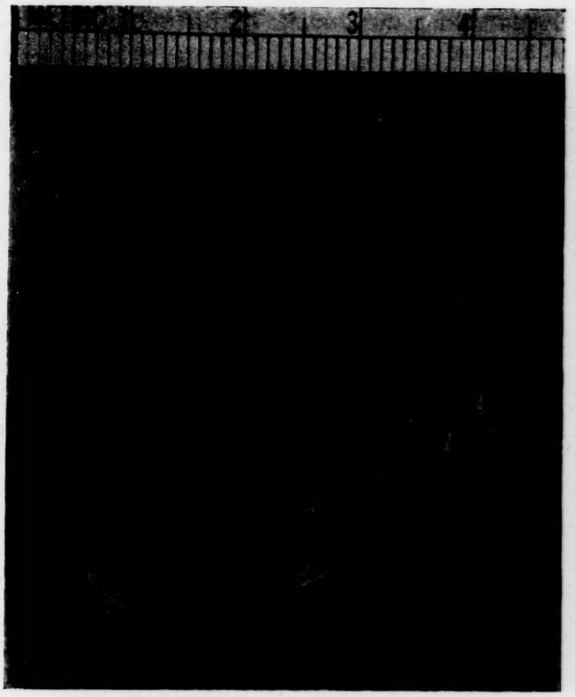

FIG. 1. Case 1. Gross section of the pheochromocytoma.

The tumor (Fig. 1) was not weighed but it measured 4 by 4 by 5 cm. Microscopically, it consisted of a fine fibrous tissue network in which lay clusters of large cells with large vesicular nuclei. The tumor was very vascular and in several areas there was a suggestion that it was growing into venous channels. The final diagnosis of the Pathology Department was, "adrenal pheochromocytoma, probably malignant."

A histamine test with 0.025 mg. histamine performed before his discharge produced no rise in blood pressure and his response to the cold pressor test at this time was remarkably small. The patient returned to work and until the time of this writing he has remained free of further attacks and has presented no evidence of metastasis.

Comment. This patient's symptoms were characteristic of paroxysmal hypertension due to discharge of adrenalin-like substances. The onset over the space of several heartbeats, his emphasis on the forcefulness rather than the rapidity of his heartbeats, the transient pounding headache, the blanching of his face, the fact that his blood pressure had been recorded during an attack at 220, compared with normal levels between attacks, are all typical manifestations of this syndrome. Most of his previous diagnoses had been in the categories of psychoneurosis. Despite

the fact that the attacks began under circumstances of stress, the patient's excellent past record, his having volunteered for service in the Armed Forces, his record of continued accomplishment after discharge and the absence of other manifestations of psychoneurosis all point

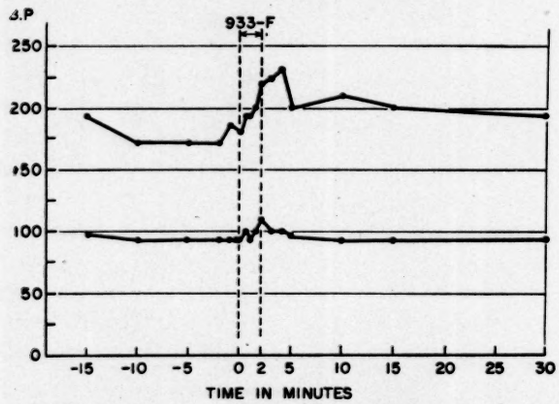

FIG. 2. Case 1. False negative benzodioxane test preoperatively. Despite elevation of blood pressure in the control period, injection of benzodioxane did not induce the expected fall in pressure but a transient greater elevation.

very strongly to his having an essentially well adjusted personality. Weiss and English[7] properly emphasize that the diagnosis of psychoneurosis should be made on the grounds of positive evidence rather than by process of exclusion. This patient's symptoms were unusual but were distinctly different from those experienced in anxiety attacks.

The other previous diagnosis, paroxysmal tachycardia, neglected the details of his attacks. Paroxysmal tachycardia consists of a run of beats from an ectopic focus, thus having a truly sudden onset in comparison with the onset over a period of several heartbeats as in this man's attacks. When questioned closely patients can make this distinction without difficulty. In contrast to the experience of Cahill and Aranow[6] this man did not experience noticeable sweating with his attacks.

The only spontaneous attack observed occurred after he had bent forward with knees straight to touch his fingertips to the floor. When he arose to the erect position his face was seen to be balanced. His blood pressure was taken immediately and found to be 256/110 and his heart sounds were found to be much louder than before. The blood pressure returned to the pre-exertion level of 180/100 in three minutes. This attack produced no symptoms. Manipulation of the adrenal areas and other types of activity on the patient's part failed to produce change in blood pressure.

Goldenberg et al.[3] have described a test for circulating adrenalin or nor-adrenalin based on the depressor, "adrenolytic" or adrenalin-

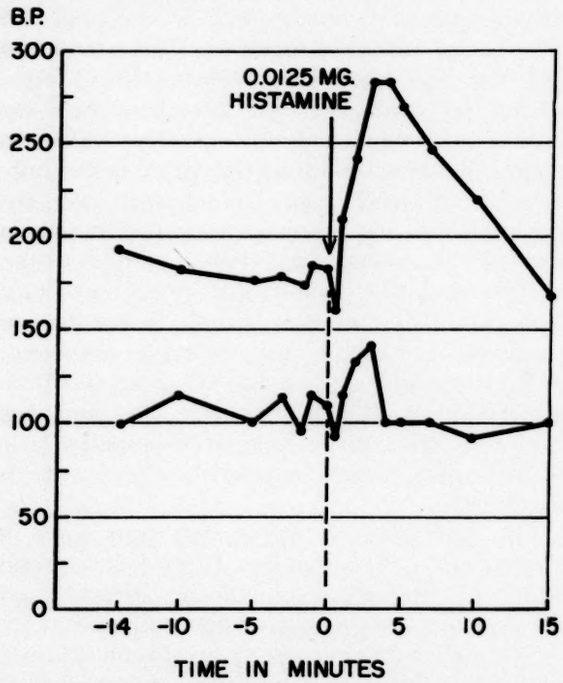

FIG. 3. Case 1. Histamine test preoperatively; typical response to intravenous injection by rapid rise of 100 mm. Hg in the systolic and 30 mm. Hg in the diastolic pressure.

inhibiting effect of benzodioxane (933F) in the presence of these substances. According to this test when blood pressure is elevated due to the circulation of pressor substances from a pheochromocytoma, the intravenous injection of benzodioxane should produce a prompt fall in blood pressure, a positive test. This test should not be expected to be informative of course unless the basal blood pressure is elevated by circulating adrenalin-like substances. If following the injection an elevated basal blood pressure does not fall, the test may be considered negative.

On the assumption that this patient's moderately elevated resting blood pressure was due to circulating adrenalin from a pheochromocytoma, a benzodioxane* test was performed, with a

* The benzodioxane used in this report was obtained through the courtesy of Dr. J. E. Howard to whom it was supplied by Merck and Company.

paradoxical rise in blood pressure rather than the expected fall. (Fig. 2.) Convinced that this was a false negative test, on a later date we injected 0.0125 mg. histamine base[2] intravenously, with the dramatic pressor response shown in Figure 3 greatly exceeding his response to the cold pressor test (Fig. 4), thus producing strong evidence of a functioning pheochromocytoma.

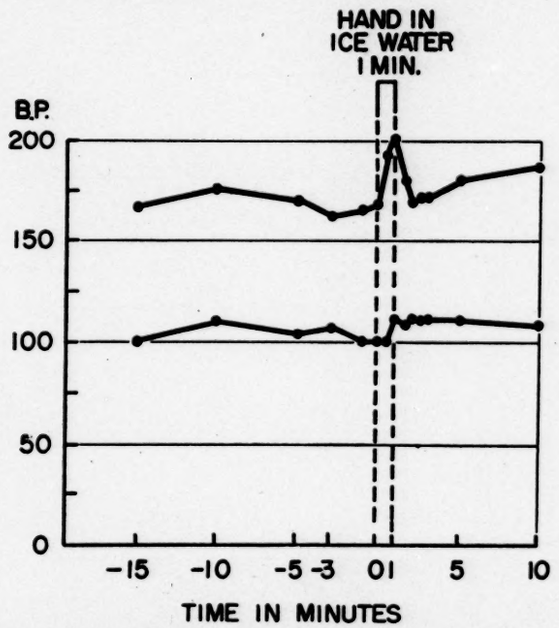

FIG. 4. Case I. Cold pressor test preoperatively; placing hand in ice water for one minute produced elevation of systolic pressure by 34 mm. Hg.

At the suggestion of Drs. J. E. Howard and Evan Calkins and after the similar experiment of Spear and Griswold[4] with dibenamine, the effectiveness of benzodioxane was clearly shown by another test in which benzodioxane injected intravenously was able to block the pressor response to a subsequent injection of histamine. (Fig. 5 compared with Fig. 3.)

After baseline recordings and intravenous injection of 20 mg. of benzodioxane as in the usual benzodioxane test, an interval of three minutes was allowed to elapse so that the immediate symptoms and effects of benzodioxane could wear off. Then 0.0125 mg. of histamine base was injected intravenously and there followed a transient drop in blood pressure of 25 mm., followed by a continuation of the gradual upward drift which occurred during the entire observation.

This response to histamine was markedly different from the response obtained in the absence of benzodioxane. We infer that benzodioxane, via its "adrenolytic" action, blocked the pressor response, since this histamine was active as evidenced by the usual initial drop in pressure, flushing and headache.

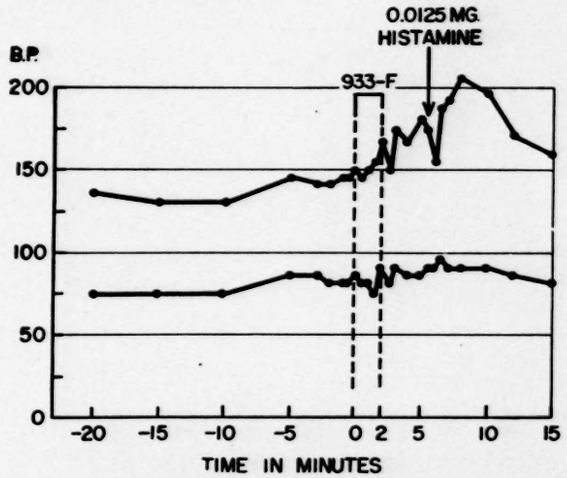

FIG. 5. Case I. Preoperative combined benzodioxane-histamine test; intravenous injection of benzodioxane was followed by slight elevation of systolic pressure. Subsequent intravenous injection of histamine evoked far less pressor response than it did in the absence of benzodioxane (compare Figure 3).

CASE II.* E. G., a twenty-eight year old housewife was admitted on January, 24, 1950, complaining of hypertension of fourteen years' duration. One brother was said to have had transient hypertension, the details of which could not be obtained. At ten years of age she had scarlet fever without apparent complication. Otherwise she had been in excellent health except for the present illness.

At fourteen years of age she was told her blood pressure was 140, at seventeen that it was 160, but there were no symptoms at that time referable to her cardiovascular system. In 1949 she became pregnant. There was an excessive weight gain in the third trimester and in the thirty-eighth week she noted several brief "crushing" headaches. Her blood pressure was found to be further elevated to 180/110 and the trace of albumin present in her urine from early pregnancy had increased to 2 plus. Examination of her retinae showed "grade I arterial spasm," with no other abnormality. She was delivered uneventfully with no hypertensive crisis, but the

* Case of Dr. E. Cowles Andrus who kindly allowed it to be reported here.

day following delivery her blood pressure rose over 220/120, accompanied by severe headache requiring an opiate for relief.

Following discharge from the hospital the severe headaches recurred intermittently. During one of these episodes she developed a central

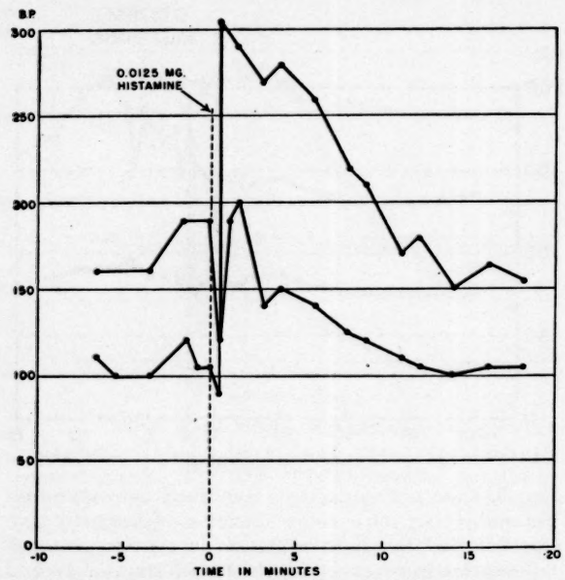

FIG. 6. Case II. Preoperative histamine test, similar to Case I.

scotoma in the left eye, and there was temporary blurring of vision in the right eye on another occasion. She described the headaches as "crushing and throbbing" and she noted considerable sweating with them. She often felt during the headache that her heart was too big for her chest and it pounded forcibly. The headaches sometimes lasted for several hours. She was referred to Dr. E. C. Andrus for investigation.

Physical examination revealed the following: Temperature 98.6°F., pulse 84, blood pressure 190/130, height 67 inches, weight 195½ pounds. She was a well developed, obese, white female who did not appear ill. There was marked spasm of the retinal arteries, with flame-shaped hemorrhages in both eyes and a large area of hemorrhage and exudate in the inferior temporal area of the left eye. The head was otherwise normal. The thyroid was palpable but normal. The lungs were clear, the heart entirely normal except for a soft systolic murmur at the base which varied with respiration. The remainder of the physical examination was not remarkable.

Laboratory data were as follows: *Blood:* corrected sedimentation rate, 11 mm./hr.; hemoglobin 15.0 gm.; hematocrit 46.0 per cent; white blood count 10,350; polymorphonuclears 51; juvenile forms 1; eosinophils 3; lymphocytes 36; monocytes 6. Cholesterol was 379 mg. per cent and non-protein nitrogen 30 to 40 mg. per cent. The basal metabolic rate was plus 5; PSP excretion 35 per cent in fifteen minutes; 75 per cent

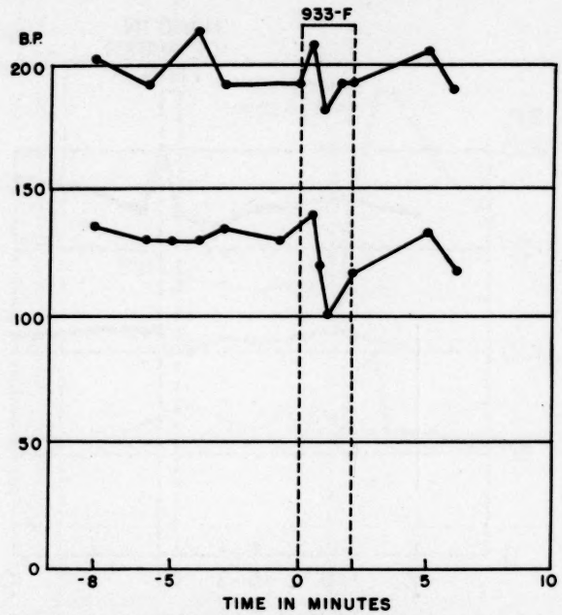

FIG. 7. Case II. False negative benzodioxane test preoperatively, similar to Case I.

in two hours. *Urinalysis:* specific gravity 1.022; negative for sugar and white blood cells; albumin 2 plus; red blood cells 5, and rare granular casts. The glucose tolerance test with the oral administration of 100 gm. of glucose revealed the following in milligrams per cent: fasting, 108; 30 minutes, 176; 1 hour, 120; 2 hours, 120; 3 hours, 80; 4 hours, 68; 5 hours, 88. The electrocardiogram was normal except for a low T_1. A chest film and intravenous pyelogram demonstrated no abnormality. Laminograms suggested a mass at the upper pole of the left kidney.

On February 6, 1950, after the diagnosis of pheochromocytoma had been established by the tests described later, Drs. W. W. and H. W. Scott, removed a tumor 3.5 by 4 cm. in size from the left adrenal area through a transverse upper abdominal incision. During manipulation of the tumor there were wide fluctuations of blood pressure. It was necessary to maintain the blood pressure during the operation and in the first twenty-four hours postoperatively by an intravenous infusion of 5 per cent glucose containing 1.5 mg. of adrenalin/100 cc.; the patient received a total of 19 mg. of adrenalin intrave-

nously in the twenty-four-hour period. The tumor histologically was a benign pheochromocytoma.

The subsequent course was entirely uneventful. The blood pressure had fallen to 120/80 at discharge, the albumin and casts disappeared from the urine and a postoperative histamine test failed to produce a significant rise in blood pressure. Follow-up examination three months after discharge revealed the blood pressure to be 120/70, with no recurrence of symptoms.

Comments. Although the presence of a pheochromocytoma was suspected at the time of admission, the symptoms in this case were not quite so classical as those in Case I. The first benzodioxane test gave equivocal results (not charted) but a Roth histamine test, using 0.0125 mg. histamine base intravenously, produced a clear-cut pressor response (Fig. 6), reproducing symptoms of a spontaneous attack. The benzodioxane test was repeated (Fig. 7); and since the initial blood pressure, averaging 200/130, failed to fall significantly after the injection of benzodioxane, the test was distinctly a false negative one. However, benzodioxane was shown to be effective to some extent in this patient on another occasion (Fig. 8) when it was injected at the height of a spontaneous attack and effected an immediate fall in pressure with remission of symptoms.

It is clear from these two cases that the benzodioxane test alone cannot be relied upon to rule out a pheochromocytoma. Obviously some factor other than circulating adrenalin maintains the hypertension in these cases which fail to respond to benzodioxane and in other cases in which the blood pressure falls slowly following removal of the pheochromocytoma. This has been discussed by Calkins and Howard.[8] Whatever the mechanism of the false negative benzodioxane test may be, the fact that it occurs is important from the diagnostic standpoint because the reliability of the benzodioxane test is thereby limited.

Another feature presented by these two cases deserves some comment, namely, that benzodioxane was able to prevent the full effects of an attack, which was histamine-induced in Case I and spontaneous in Case II. On reviewing the fatal operative cases in the literature it is apparent that some patients die from pressor withdrawal shock, preventable as in Case II by injections of adrenalin intravenously. Other patients[9,10] suffer from "shock" which appears to result from excess release of pressor substance with such severe constriction of the peripheral arteries that blood pressure becomes unrecordable. It might be possible to prevent this adrenalin shock as well as the more common complications of sudden severe hypertension by the judicious use of benzodioxane at the time a pheochromocytoma is manipulated prior to ligation of its blood supply. The combined benzodioxane-histamine test as used in Case I may prove capable of predicting the usefulness of benzodioxane in this way.

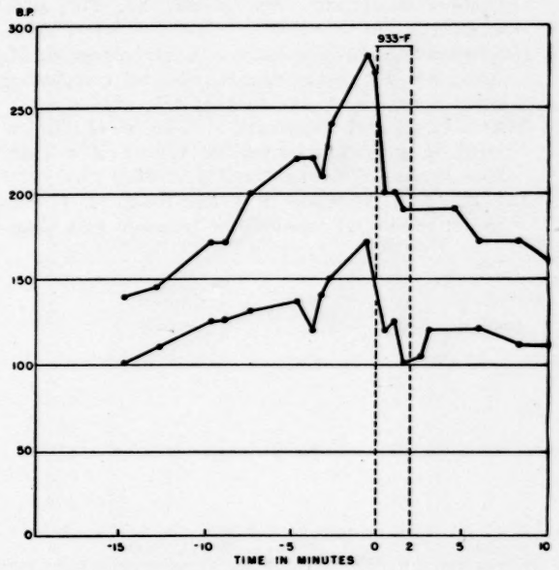

FIG. 8. Case II. Effectiveness of benzodioxane demonstrated by termination of a spontaneous attack preoperatively.

SUMMARY

1. The history may be sufficiently typical in a case of pheochromocytoma to warrant exploration without further tests or in the face of equivocal tests.

2. Laminography without perirenal air insufflation was an effective means of locating the tumor in these cases.

3. These patients were proven to have false negative benzodioxane tests for pheochromocytoma. This restricts the reliability of this substance as a diagnostic agent in this condition unless it is used in combination with other tests.

4. Intravenously injected benzodioxane (933F) has been shown to block the pressor response to subsequent histamine injection in a patient with pheochromocytoma. The possible therapeutic use of this test is discussed.

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Rupture of Abdominal Aneurysm into the Gastrointestinal Tract*

ELI ANTZIS, M.D., JAMES DUNN, M.D. *and* ANTHONY J. SCHILERO, M.D.
New York, New York

RUPTURE of an abdominal aneurysm into the gastrointestinal tract is of sufficiently rare occurrence to justify reporting. Including the case recorded in this article a total of fifty-one cases have been reported. In a review of the subject Rottino in 1943 found thirty-one cases in the literature including one of his own. In 1946 Hunt and Weller collected ten additional cases, one of which was their own. The authors are now adding a case in addition to eight cases not tabulated previously. The cases in which adequate information was available have been reviewed.

TABLE I
INCIDENCE OF ABDOMINAL ANEURYSM IN RELATION TO ETIOLOGY AND SEX

Author	Etiology	Percentage	Ratio Male: Female
Scott[20] (94 cases)	Syphilis	59.6	5.8:1
	Arteriosclerosis	21.3	
	Mycosis	19.1	
Mills and Horton (80 cases)	Syphilis	8.8	2:1
	Arteriosclerosis	86.2	
	Mycosis	2.5	
Kampmeier[12] (68 cases)	Syphilis	57	5:1
Hubery and Pollack (48 cases)	Syphilis	75	4:1
	Arteriosclerosis	17	
Blakemore[5] (32 cases)	Syphilis	19	5:1
	Arteriosclerosis	81	

Etiology. Syphilis and arteriosclerosis are the two most common causes of aneurysm of the abdominal aorta. Other etiologic factors are trauma, tuberculosis and infection. Syphilis was the predominant factor in the younger age groups whereas arteriosclerosis was predominant in older age groups. We believe that the marked variations in incidence reported by different authors are due to the following reasons: (1) whether hospital-handled, acute or chronic cases; (2) whether syphilis was relatively common in patient populations studied. (Table I.)

Age and Sex. In a review of fifty-one cases in which aneurysm of the abdominal aorta had ruptured into the gastrointestinal tract, the average age was found to be 52.5 years (oldest eighty-one, youngest twenty). There were forty-one males and ten females, a ratio of approximately 4:1. Aneurysms due to arteriosclerosis usually ruptured from the age of forty on, luetic aneurysm more commonly from ages twenty to sixty, mycosis and tuberculosis in younger and middle-aged groups. (Table II.)

TABLE II*
AGE GROUPS IN RELATION TO ETIOLOGY

Age	Arteriosclerosis	Syphilis	Mycosis	Tuberculosis
20–30		1	3	
31–40		3		1
41–50	2	3	1	
51–60	5	2	1	
61–70	2	1		
71–80	6			

* Full information on all cases reviewed was unavailable because of incomplete reports.

Symptoms and Signs of Abdominal Aneurysm. The presenting symptoms may be pain in the abdomen and back which are referred to the hip, leg and scrotum. Pressure symptoms on the vertebrae are characterized by an articular-type pain in the back; on the gastrointestinal tract by indigestion, nausea, anorexia, intestinal obstruction, loss of weight, vomiting; and on the genitourinary tract by oliguria and anuria. A pulsating, expansile, abdominal mass (Table III) with thrill, murmur and bruit may be present. Surprisingly enough, in only five of the forty-seven cases reviewed was there a thrill, murmur, bruit or combination of these found.

According to Kampmeier, the basic clinical picture of abdominal aortic aneurysm is associated with pressure of abdominal tissues in 60 per cent of the cases and expansile pulsation in 98 per cent of these. A palpable tumor and roentgenographic evidence of calcified ab-

* From the Second Medical Division of the Goldwater Memorial Hospital, Department of Hospitals, City of New York, N. Y.

dominal mass, with external erosion on an indefinite soft tissue mass, may be evident. Lipschutz and Chodoff[15] added to the general picture of ruptured abdominal aortic aneurysm by including a vascular axis and a state of shock, high leukocyte count and moderate elevation of diastase content of urine. Lang[14] stated that four of five arteriosclerotic aneurysms are asymptomatic while four of five luetic aneurysms produce symptoms.

TABLE III
EXPANSILE PULSATILE MASS—THIRTY-ONE CASES

Thrill	Murmur	Bruit
2	3	2

TABLE IV
TIME INTERVAL BEFORE DEATH

Death	Cases
Rapidly	9
Few minutes	1
40 minutes	1
1 day	4
2 days	4
3 days	1
4 days	1
5 days	1
7 days	3
8 days	3
9 days	1
2 weeks	1

Thus we can now summarize the symptoms of abdominal aortic aneurysm as follows: (1) pulsatile expansile abdominal mass, (2) arteriosclerotic thrill, murmur, bruit, (3) erosion of lumbar vertebrae, (4) soft tissue mass, (5) calcification in wall of aneurysmal sac, (6) displacement of viscera with or without symptoms and (7) radicular referred pain. The syndrome of rupture of an aneurysm of the abdominal aorta into the gastrointestinal tract may be characterized by sudden hematemesis, melena and shock. (Tables IV, V and VI.)

The diagnosis of the case presented by the present authors was made on the basis of an expansile, pulsatile abdominal mass (no thrill, bruit or murmur), sudden episode of hematemesis, shock and melena and a negative history or clinical course suggestive of gastrointestinal ulcer or malignancy.

CASE REPORT

The patient was a seventy-eight year old colored female who was first admitted to the Goldwater Memorial Hospital in January, 1949, from an old age home because of fever of 102.4°F. and semi-stupor. At other hospitals the patient had previously been diagnosed as having generalized arteriosclerosis, arteriosclerotic and hypertensive heart disease with auricular fibrillation. On admission, examination revealed a poorly nourished Negress, semi-stuporous with engorged neck veins, emphysematous chest, diminished breath sounds at both bases, auricular fibrillation, enlarged heart to the left and blood pressure 150/100. Abdominal examination was negative. Laboratory findings included white cell count 12,000, blood urea nitrogen 39.4 mg. per cent, negative blood Mazzini test. The remainder of laboratory tests were within normal limits. Chest x-ray revealed enlarged heart to the left and congestion of the lung fields. The patient was treated with penicillin, digitalis, mercurial diuretics and salt-free diet. Following this regimen the temperature fell to normal; blood urea nitrogen became 13 mg. per cent. The patient was discharged two and a half months after admission.

TABLE V
ORGAN INVOLVED IN RUPTURE

Site of Rupture	Cases
Duodenum	36
third portion	32
second portion	2
unspecified	2
Stomach	7
Jejunum	5
Small intestine	1

TABLE VI
CORRELATION OF SITE OF RUPTURE OF ANEURYSM AND ORGAN AFFECTED

	Duodenum			Undetermined	Stomach	Jejunum
	1st	2nd	3rd			
Celiac axis						
Above	..	1	..	..	..	..
At level of	..	..	..	3	2	1
Superior mesenteric						
Above	..	..	1	..	..	..
At level of	..	..	1	1	..	..
Below	..	..	4	1	..	1
Renal arteries						
Above	..	..	1	..	..	..
Below	..	..	9	1	..	1
Inferior mesenteric						
Above	..	..	1	..	1	..
Below	..	..	1	..	..	..
Just above bifurcation	..	..	..	1	..	2

Five months later in August, 1949, the patient was readmitted for progressive breathlessness, weakness, precordial distress and palpitations, more marked during the preceding two weeks,

An accurate history could not be obtained. Temperature was 100°F., pulse 63 (irregular), respirations 18 and blood pressure 160/90. Examination disclosed a poorly nourished female appearing chronically ill, depressive and with apparent memory defects. Neck veins were distended, and diminished breath sounds, medium moist rales, dullness and diminished tactile fremitus were noted in left posterior base. The heart was enlarged with apex beat in the sixth intercostal space in the anterior axillary line. A forceful thrust was noted with a harsh systolic murmur at apex and base. P_2 was greater than A_2, heart rate 100 and grossly irregular. The liver was felt three fingers below the costal margin but the kidney and spleen were not palpable. There was slight pretibial edema bilaterally but no clubbing or cyanosis of digits. Gradual improvement in symptoms occurred with mercurial diuretics, intermittent digitalis, salt-free diet and oxygen. Laboratory data revealed 12.5 gm. per cent hemoglobin, 6,750 white cells, 80 per cent polymorphonuclears, 16 per cent lymphocytes, 8 per cent monocytes; blood urea nitrogen 21.5 mg. per cent; blood sugar 68 mg. per cent; blood Mazzini test negative; sedimentation rate 5 mm. in the first hour, hematocrit 38 per cent. Chest x-ray revealed no change from previous admission. Electrocardiograph showed left axis deviation, rate of 100, intermittent flutter-fibrillation and multiple premature ventricular contractions. The patient improved. On December 1, 1949, it was noted that the patient had a sausage-shaped mass in the right upper quadrant extending from the umbilicus to just below the costal margin to the right of the midline. Expansile pulsation was present but no bruit, thrill, murmur or tenderness was found. The cardiac status remained improved and all signs of peripheral edema and enlarged liver had disappeared.

On February 5, 1950, at 1:30 P.M., the patient was found lying on the floor of the bathroom in a semi-conscious state. She was carried to bed and in the next five minutes she vomited 900 cc. of bright red blood mixed with gastric contents. She complained of no pain but did feel dizzy and nauseous. The pulse was imperceptible; heart sounds could not be heard and blood pressure was not obtainable. The lungs were clear. A non-tender pulsatile mass was present in the right upper quadrant. Blood count at this time revealed 3,000,000 red cells, 12,200 white cells, 8 gm. per cent hemoglobin, 80 per cent polymorphonuclears, 18 per cent lymphocytes and 2 per cent eosinophils. Following infusions of plasma, glucose and whole blood, the heart sounds and pulse became stronger. Peristaltic sounds were no longer heard in the abdomen. The blood pressure rose to 126/60 eleven hours following the acute episode but then the patient vomited 3,000 cc. of bright red blood mixed with clots, the pulse became rapid, thready and irregular and the blood pressure dropped to 80/60. An additional 1,500 cc. of whole blood, 3 units of plasma and infusions of glucose were given with resultant rise of red cells to 4.1 million, hemoglobin 9.5 gm. per cent and hematocrit 38 per cent.

Her course was progressively downhill; grossly bloody stools were passed, respirations became rapid, moderate spasticity was noted in the right upper quadrant with moderate tenderness over the pulsating mass and the skin became cold and clammy. The patient expired thirty-one hours after onset of hematemesis. The antemortem diagnosis was abdominal aortic aneurysm with rupture into upper gastrointestinal tract.

Microscopically, section through the wall of the aorta revealed the intima to be completely hyalinized. Its inner half, however, was the seat of extensive calcific deposits many areas of which appeared to be transforming into cartilage. Other portions of the inner half of the media were spottily calcified while the major portion of this section was apparently necrotic and contained many clefts left by dissolved cholesterol crystals. Occasional portions of the intima appeared normal while others were thickened by hyalinized connective tissue. Surrounding certain calcific deposits were patchy areas of cellular infiltration composed of round cells, a few plasma cells and histiocytes together with small vasa vasorum. Sections through the aneurysmal wall showed an old hyalinized clot adherent to the intima and merging imperceptibly into the arterial wall. In this area many newly formed capillaries appeared to extend from the intima forward and into the clot. The media in areas through the aneurysmal sac was thinned out, its muscular and elastic fibers were replaced by hyalinized connective tissue. Where the muscle and elastic fibers of the media were still intact, the width of this section was markedly diminished. At the junction of media-adventitia there were areas of necrotic connective tissue containing lymphocytes, histiocytes, occasional plasma cells and hemosiderin-containing macrophages. The adventitial connective tissue was also hyalinized to a great extent and occasion-

FIG. 1. Aortic abdominal aneurysm with perforation into third part of duodenum, showing lumen of aneurysm.

ally contained a collection of lymphocytes which were not perivascular. The aneurysmal wall contained very little remaining adventitia. Sections through this area showed the thinned out adventitia closely applied to lymphatic tissue. The over-all thickness of the sac wall was approximately half that through the remainder of the aortic wall.

The ascending aorta and descending thoracic aorta presented a moderate degree of intimal atherosclerosis. In the abdominal aorta, below the level of the renal vessels, there was a saccular aneurysmal dilation of the vessel, the size of an egg, which contained some old adherent blood clot. The wall of the aneurysm presented marked atherosclerosis. Viewed anteriorly the aneurysm appeared as a bluish mass which underlay the proximal portion of the superior mesenteric artery and appeared to include the inferior mesenteric artery within the sac. However, there was no infarction of the bowel nor any retroperitoneal hemorrhage. An opening about 1.0 to 1.5 mm. in diameter was noted between the aneurysm and the third portion of the duodenum which was adherent to the sac. There was no blood in the stomach or duodenum. The large bowel, beginning at the cecum, was moderately dilated and bluish black due to the presence of dark red blood within the hernia. (Fig. 1.)

Of interest is the fact that in this case, as in previously reported cases with the exception of one reported by Coggeshall and Genovese,[7] there was a direct communication between the aneurysm and the gastrointestinal tract.

SUMMARY AND CONCLUSION

1. A review of the literature from 1843 reveals fifty-one reported cases of perforation of abdominal aorta into the gastrointestinal tract. These have been reviewed and analyzed in respect to etiologic factors, sites of perforation, symptomatology and clinical findings.

2. Another case of rupture of abdominal aneurysm into the third portion of duodenum is recorded.

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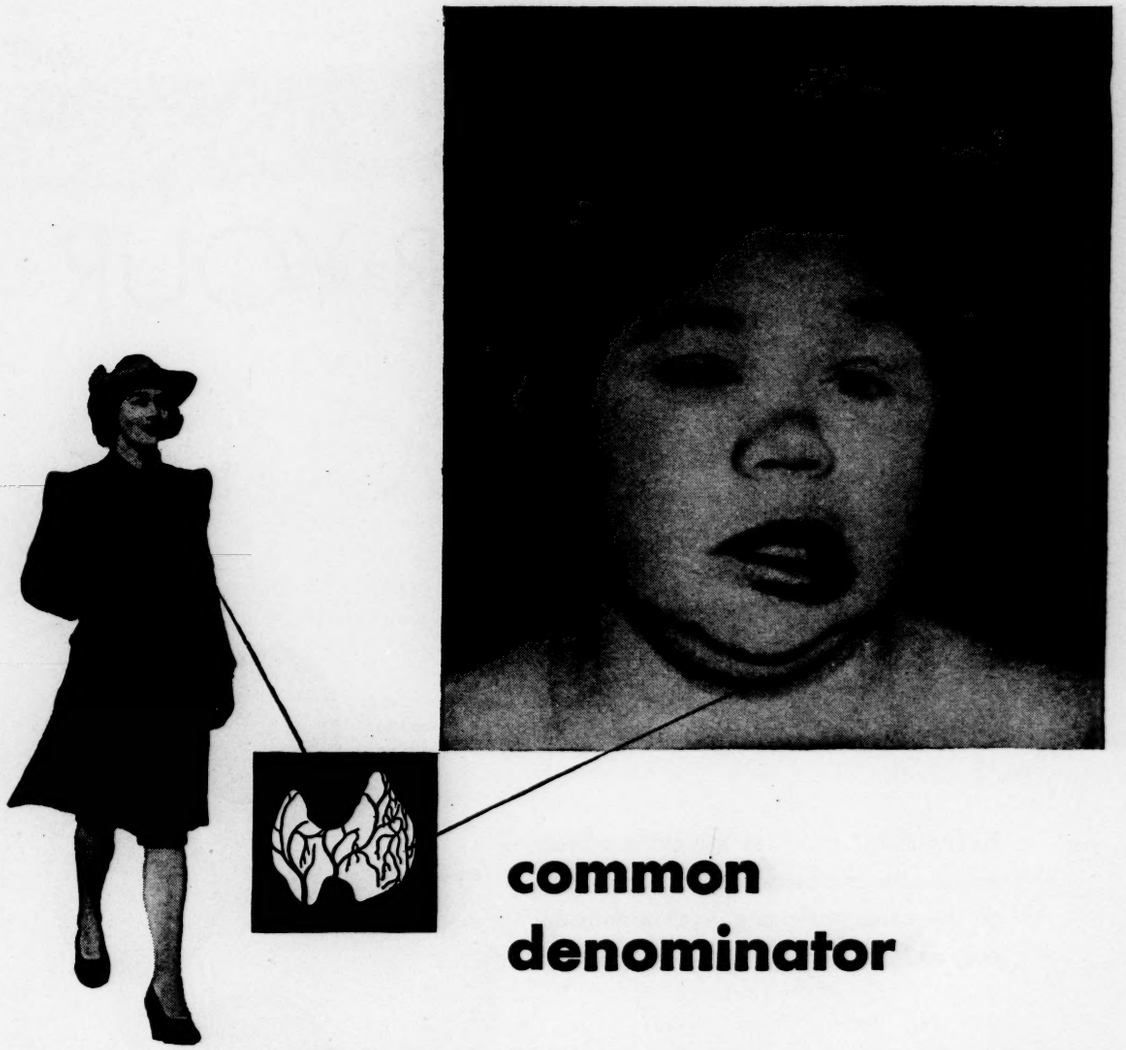

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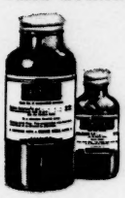

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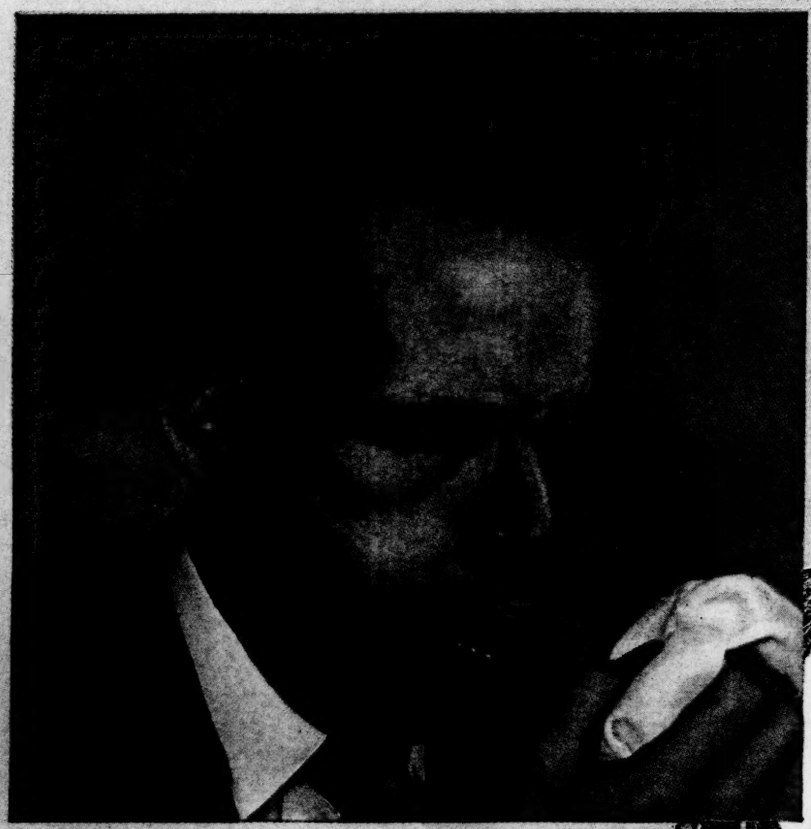

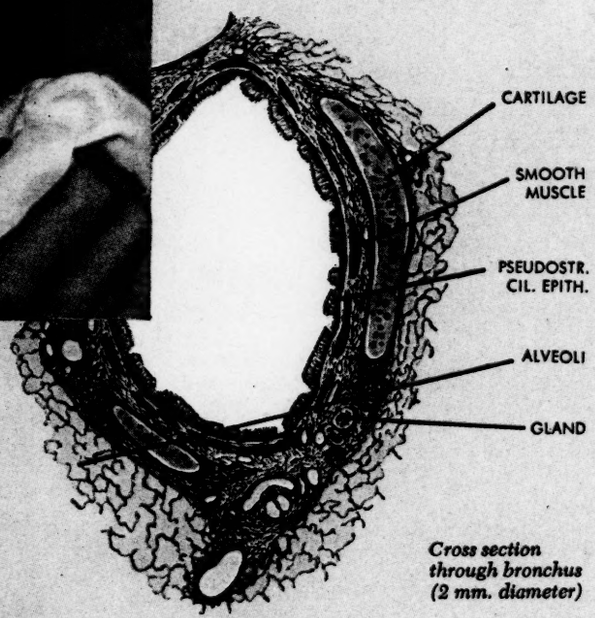

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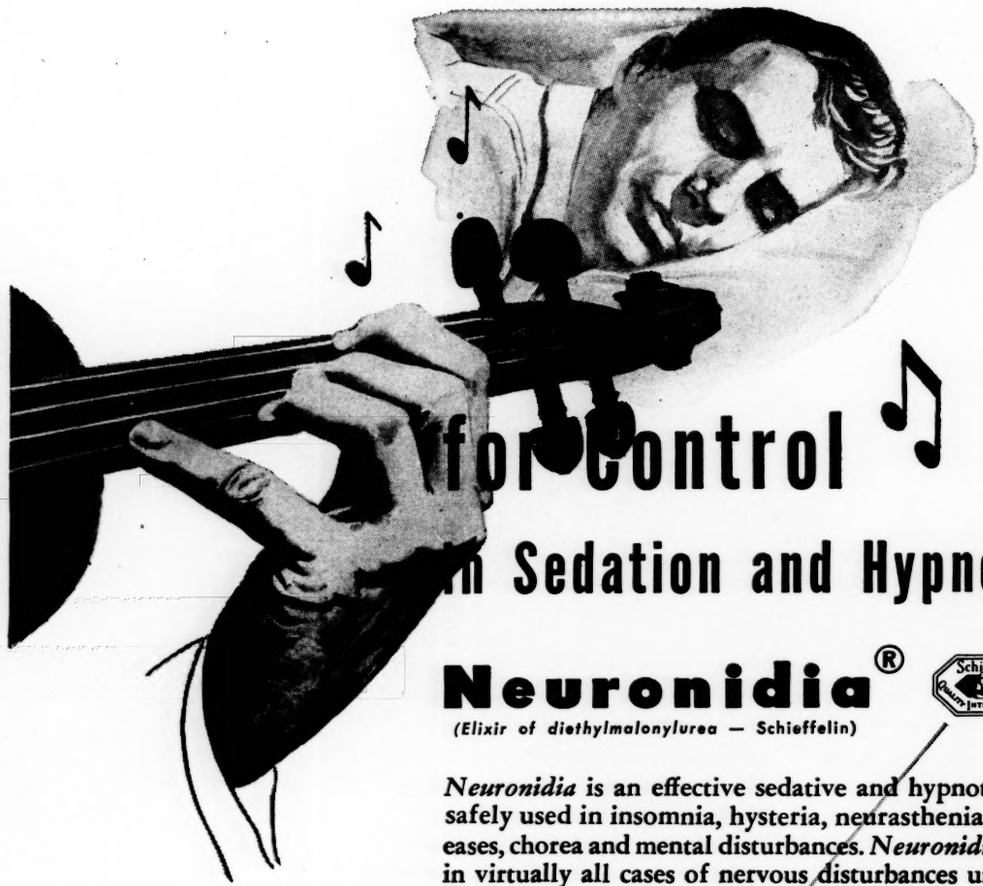

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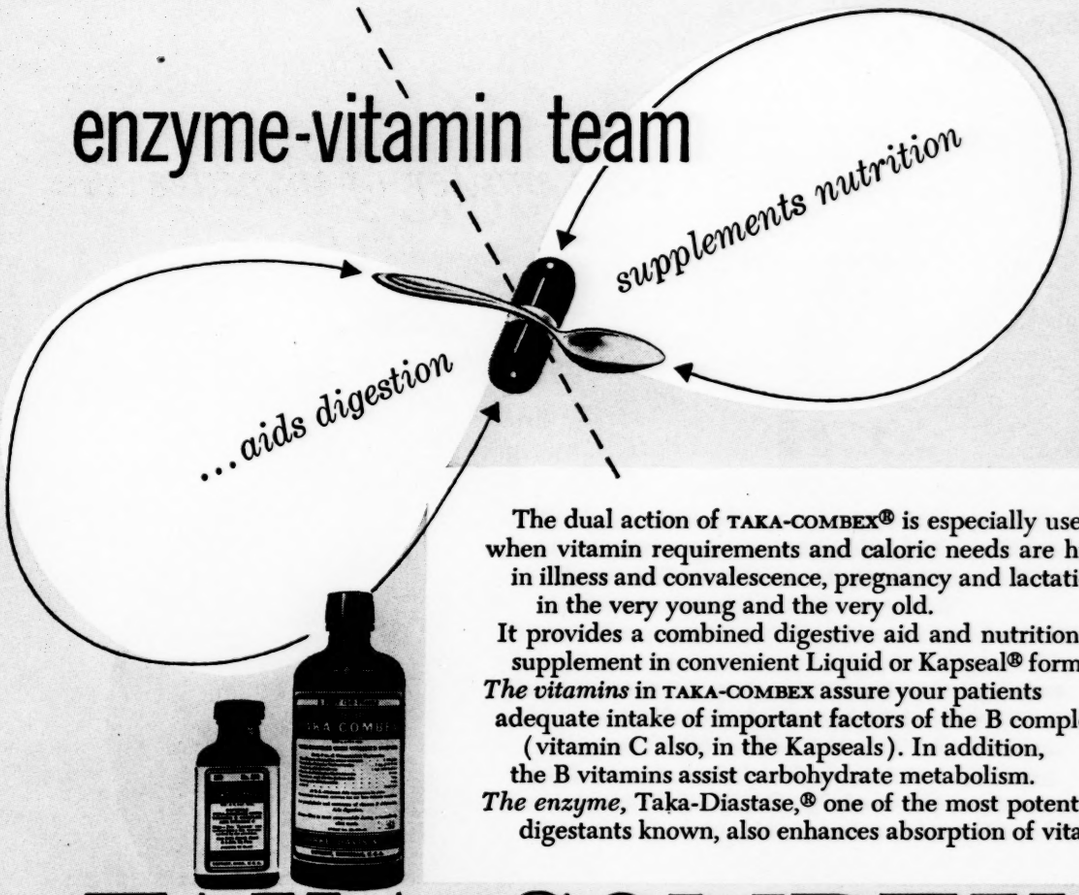

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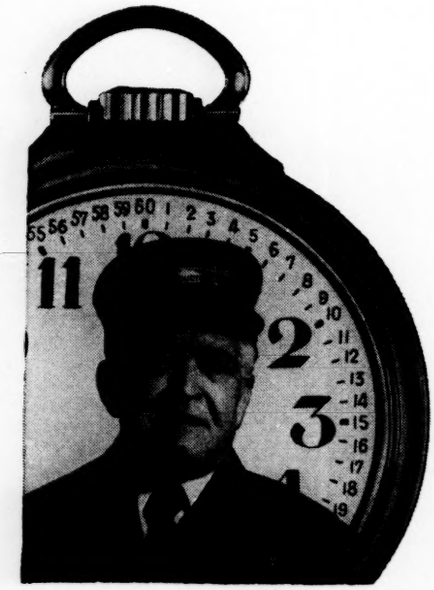

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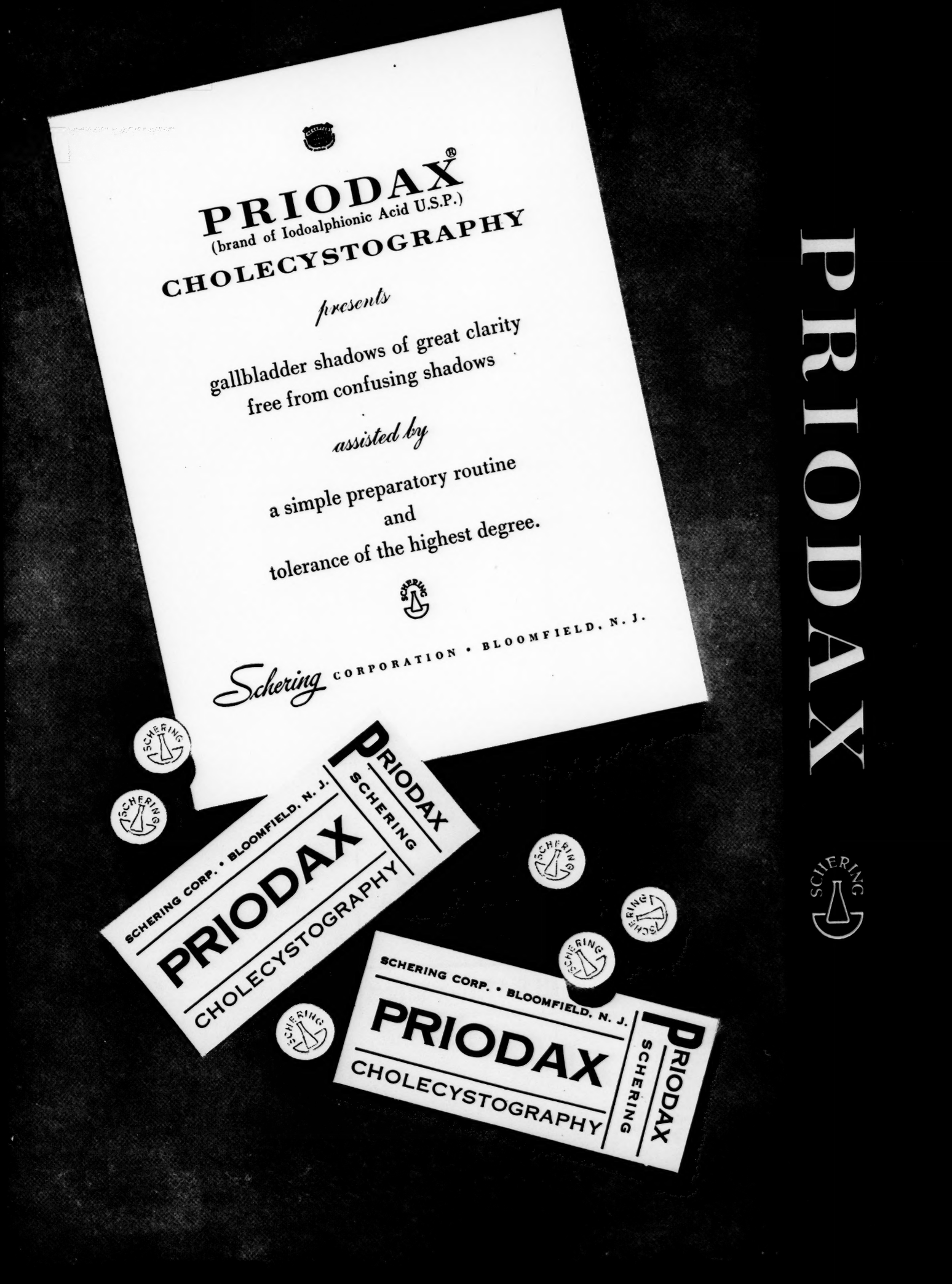
PRIODAX®
(brand of Iodoalphionic Acid U.S.P.)
CHOLECYSTOGRAPHY
presents
gallbladder shadows of great clarity
free from confusing shadows
assisted by
a simple preparatory routine
and
tolerance of the highest degree.
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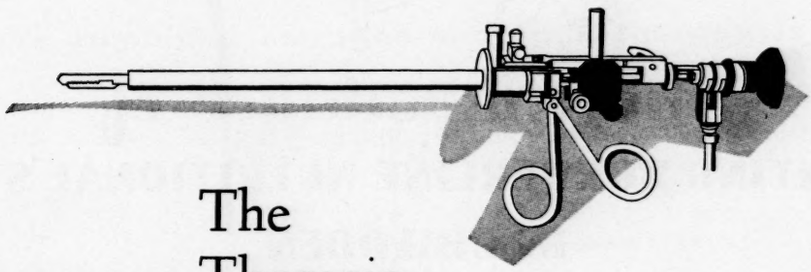
AUREOMYCIN

Hydrochloride Crystalline

Effective against many bacterial and rickettsial infections, as well as certain protozoal and large viral diseases.



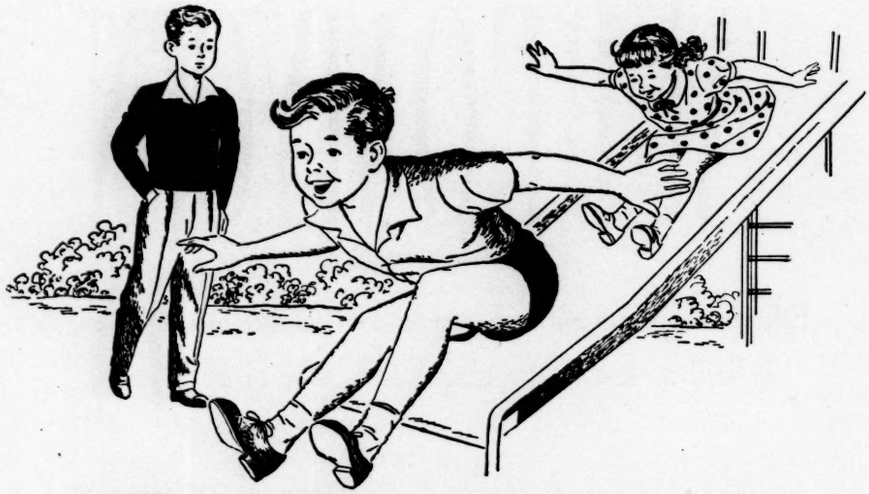
The Thoracic Surgeon . . .

will find aureomycin of benefit in the complications of pneumonias refractory to other forms of therapy, particularly in those very serious forms caused by the staphylococcus or by *Klebsiella pneumoniae.* In the pneumonic involvements of psittacosis, tularemia, rickettsial disease or mucoviscidosis, aureomycin is highly effective. It is also very useful in the ambulatory or surgical management of bronchiectasis. Multiple lung abscesses have been known to heal with aureomycin treatment alone. In operative thoracic procedures, aureomycin is invaluable.

Packages

Capsules: Bottles of 25 and 100, 50 mg. each capsule. Bottles of 16 and 100, 250 mg. each capsule.
Ophthalmic: Vials of 25 mg. with dropper; solution prepared by adding 5 cc. of distilled water.

LEDERLE LABORATORIES DIVISION
AMERICAN Cyanamid COMPANY
30 Rockefeller Plaza, New York 20, N. Y.



The Importance of
PREVENTING BORDERLINE NUTRITIONAL STATES IN CHILDREN

IN recent years increasing interest has been focused on the relationship between nutrition and the physical, mental and emotional development of children. It is now well recognized that listlessness and apathy in the child frequently may be nothing other than manifestations of a borderline nutritional state resulting from faulty food selection and inadequate consumption. Moreover, such sequelae of faulty nutrition often respond dramatically to improved food habits.*

For preventing borderline nutritional states in children due to food whims, poor choice of foods, or lack of interest in eating, Ovaltine in milk enjoys long-established usefulness. Its rich content of biologically complete protein, vitamins and minerals can supplement even grossly deficient diets to optimal nutrition. The delicious flavor of Ovaltine invites its acceptance and lends interest to eating when the appetite lags. Children particularly like Chocolate Flavored Ovaltine.

Three servings of Ovaltine in milk furnish the supplementary amounts of nutrients shown in the appended table.

*Baumgartner, L.: Wider Horizons for Children; The Mid-century White House Conference and Children's Nutrition, J. Am. Dietet, A. 27:281 (Apr.) 1951.

THE WANDER COMPANY, 360 N. MICHIGAN AVE., CHICAGO 1, ILL.

Ovaltine

Three servings of Ovaltine, each made of ½ oz. of Ovaltine and 8 oz. of whole milk,* provide:

PROTEIN	32 Gm.	VITAMIN A	3000 I.U.
FAT	32 Gm.	VITAMIN B_1	1.16 mg.
CARBOHYDRATE	65 Gm.	RIBOFLAVIN	2.0 mg.
CALCIUM	1.12 Gm.	NIACIN	6.8 mg.
PHOSPHORUS	0.94 Gm.	VITAMIN C	30.0 mg.
IRON	12 mg.	VITAMIN D	417 I.U.
COPPER	0.5 mg.	CALORIES	676

***Based on average reported values for milk.**

Two kinds, Plain and Chocolate Flavored. Serving for serving, they are virtually identical in nutritional content.

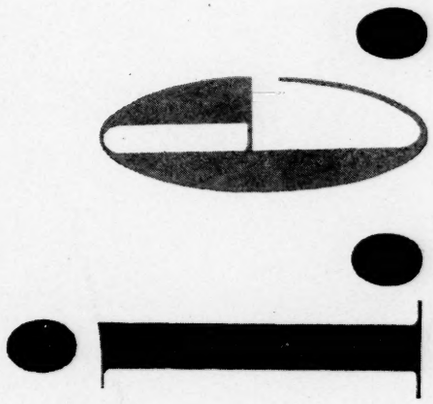
"PREMARIN"

WITH

METHYLTESTOSTERONE

i.e.

wherever combined estrogen-androgen therapy is indicated



i.e. in fractures and osteoporosis in either sex to promote bone development, tissue growth, and repair.

i.e. in the female climacteric in certain selected cases.

i.e. in dysmenorrhea in an attempt to suppress ovulation on the basis that anovulatory bleeding is usually painless.

i.e. in the male climacteric to reduce follicle-stimulating hormone levels.

"Premarin"® with Methyltestosterone

for combined estrogen-androgen therapy

A steroid combination which permits utilization of both the complementary and the neutralizing effects of estrogen and androgen when administered concomitantly. Thus certain properties of either sex hormone may be employed in the opposite sex with a minimum of side effects.

Availability: Each tablet provides estrogens in their naturally occurring, water-soluble, conjugated form expressed as sodium estrone sulfate, together with methyltestosterone.

No. 879—Conjugated estrogens equine ("Premarin").....1.25 mg.
Methyltestosterone .....10.0 mg.
Bottles of 100 tablets (yellow)

No. 878—Conjugated estrogens equine ("Premarin").....0.625 mg.
Methyltestosterone .....5.0 mg.
Bottles of 100 tablets (red)

Ayerst, McKenna & Harrison Limited
22 East 40th Street, New York 16, N. Y.

to equal
the ascorbic acid content
of "Beminal" *Forte*
with Vitamin C.
One capsule provides
100 mg., the equivalent of
at least 24 ounces
of canned tomato juice.
This is but one feature of
"Beminal" *Forte* with Vitamin C
which also contains therapeutic amounts
of important B complex factors.
(See formula below)

Ayerst,
McKenna &
Harrison
Limited

No. 817: Each dry powder capsule contains:

Thiamine HCl (B_1)	25.0 mg.
Riboflavin (B_2)	12.5 mg.
Nicotinamide	100.0 mg.
Pyridoxine HCl (B_6)	1.0 mg.
Calc. pantothenate	10.0 mg.
Vitamin C (ascorbic acid)	100.0 mg.

Supplied in bottles of 30, 100, and 1,000.

22 East 40th Street, New York 16, N. Y.

Complete and lasting relief to 90% of patients with

nausea and vomiting of pregnancy

A recent clinical study[1] finds 'Dexedrine' remarkably effective in the treatment of the nausea and vomiting of pregnancy.

The author states:

1. "In a series of 165 patients with nausea and vomiting of pregnancy, 'Dexedrine' Sulfate produced complete relief in 148, or 90% .... Marked improvement occurred in almost every case within three days .... Complete relief occurred in four to ten days."

2. "'Dexedrine' has definite advantages over other treatments, most important of which are the mental and physical alertness, and the general feeling of well-being which it produces."

The study concludes: 'Dexedrine' "usually gives prompt and lasting relief; it is effective orally; it produces no significant side effects; and it gives mental and physical stimulation which improves the patient's morale and enables her to carry on normal activities."

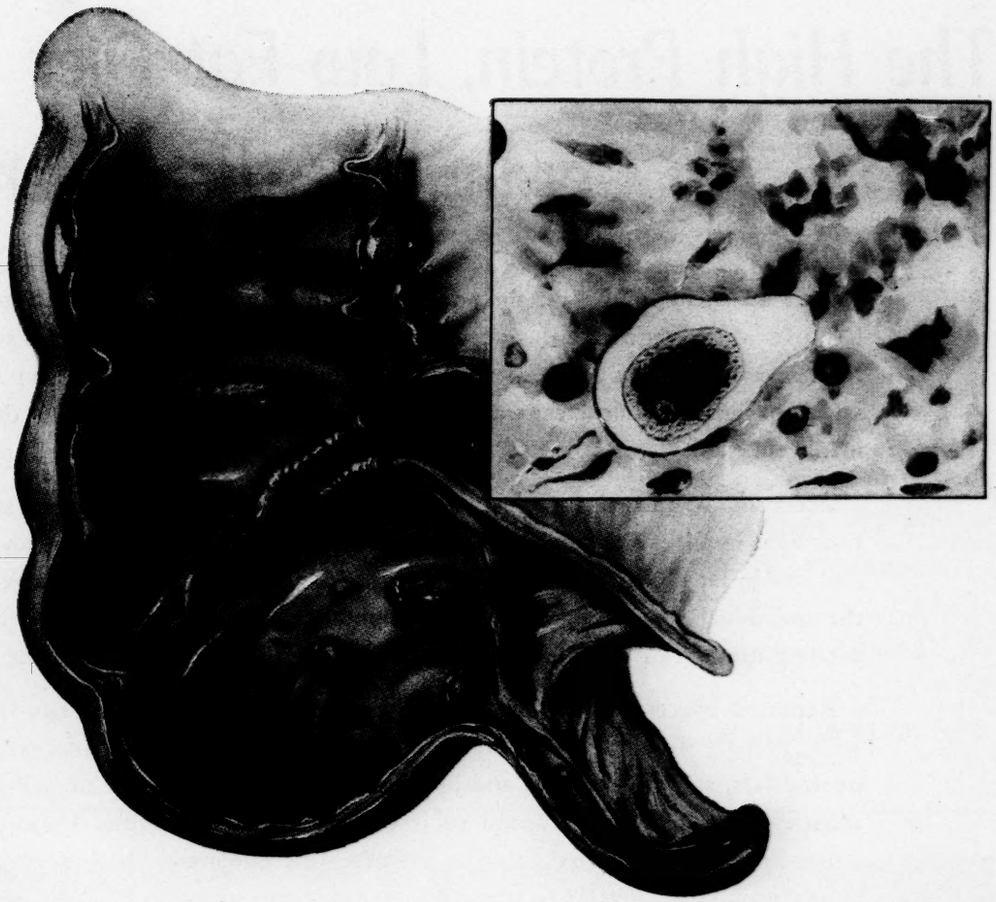
Smith, Kline & French Laboratories, Philadelphia

Dexedrine* tablets & elixir

the antidepressant of choice and the most effective drug for control of appetite in weight reduction

*T.M. Reg. U.S. Pat. Off.

1. Anspaugh, R. D.: Effects of Dexedrine Sulfate on Nausea and Vomiting of Pregnancy, Am. J. Obst. & Gynec. 60:888 (Oct.) 1950.



in amebiasis:

"In hospitals and institutions where the incidence of amebiasis is significant, simultaneous mass treatment by terramycin can control the infection . . . stool examinations performed 6 months after terramycin therapy revealed an apparent effectiveness of 100 per cent."

Most, H.; Tobie, J. E.; Bozicevich, J., and Reardon, L. V.: cited in Pub. Health Rep. 65:1684 (Dec.) 1950.

CRYSTALLINE TERRAMYCIN HYDROCHLORIDE

available | *Capsules, Elixir, Oral Drops, Intravenous, Ophthalmic Ointment, Ophthalmic Solution.*

ANTIBIOTIC DIVISION Pfizer **CHAS. PFIZER & CO., INC.,** *Brooklyn 6, N. Y.*

The High Protein, Low Fat Diet in Coronary Atherosclerosis

Recent advances in the dietary and medical management of atherosclerosis have improved the prognosis of this degenerative vascular disease. No longer is atherosclerosis regarded merely as a progressive disease of advancing age with an inevitably fatal termination, but rather as a metabolic abnormality subject to therapeutic management.

According to a recent four year study,* it is indicated that the course of atherosclerosis as it involves the coronary vessels may be favorably influenced by an adequate diet low in fat (20 to 25 Gm.), high in protein, and adjusted to the caloric needs of the individual for maintaining ideal weight. Lean meats, at both the noon and evening meals, can contribute importantly to the protein adequacy of the diet.

Reported effects of this diet included reduction in the mortality rate of patients with coronary thrombosis and myocardial infarction, reduction in total serum lipids, neutral fats, and chylomicron and lipomicron counts. An increase in well-being and capacity for work in the clinical subjects was also demonstrable. These effects, in part, were attributed to reduction in weight in the obese and "high normal" weight subjects, to psychotherapeutic results, to adequate vitamin A intake ensured by addition of a vitamin A concentrate to the diet, or to metabolic changes induced by a decreased fat intake.

Meat, a favorite protein-rich food, can contribute valuably to the protein needs of the atherosclerotic patient as well as to the normal individual. The protein of meat contains all the essential amino acids in proportions required for good growth and upkeep of tissues and for the maintenance of tissue functioning and general resistance to infection. Meat is also a rich source of iron and of vitamin B complex, including niacin, pyridoxine, riboflavin, thiamine, and the newly discovered vitamin B_{12}.

---

*Morrison, L. M.: Arteriosclerosis: Recent Advances in the Dietary and Medicinal Treatment, J.A.M.A. *145*:1232 (Apr. 21) 1951.

The Seal of Acceptance denotes that the nutritional statements made in this advertisement are acceptable to the Council on Foods and Nutrition of the American Medical Association.

In Pregnancy and Lactation

DICALOSSA TABLETS AND CAPSULES

A mineral nutritional supplement supplies those mineral deficiencies of greatest incidence.

Six tablets or capsules daily supply Dicalcium Phosphate 25.50 grs., Calcium Gluconate 20.40 grs., Exsiccated Ferrous Sulphate, U.S.P. 8.04 grs., Vitamin D (Viosterol) 2,000 U.S.P. units.

Descriptive literature and specimens available

Tablets and capsules in bottles of 100 and 1,000 (tablets in 200s also)

G. W. Carnrick Co.
NEWARK 1, NEW JERSEY

new!

GENTEXIN

contains ***sodium gentisate*** *unusual and superior* **analgesic**
more effective, better tolerated than aspirin[1]

GENTEXIN is destined to replace aspirin for rapid, non-toxic relief from pain in **headache, dysmenorrhea, neuralgia, arthralgia, myalgia, colds** and other conditions with mild to moderately severe pain.

Since untoward reactions from sodium gentisate are "unusual and insignificant," **GENTEXIN** is especially valuable in patients with chronic pain or who cannot tolerate aspirin.

Prescribe **GENTEXIN** in bottles of 50, 100 and 500 tablets.

[1] Batterman, R. C.; Kryle, H., and Dann, S.: In press, N.Y. State J.M.

GENTEXIN
elevates mood

In addition to Sodium Gentisate (200 mg.) in each tablet, **GENTEXIN** provides Acetophenetidin (200 mg.) as an analgesic potentiator, and dl-Desoxyephedrine (1 mg.) to elevate the mood which is usually depressed in painful conditions.

samples of* GENTEXIN *and literature upon request

CROOKES LABORATORIES, INC. Crookes 305 EAST 45 ST., NEW YORK, N.Y.

measuring rod for mercurial diuretics

Determining the relative clinical efficacy of mercurial diuretics by comparing the diuretic response obtained with similar doses is not entirely sound.[1] A more valid comparison is given by study of dosage-response curves[1] which show the differences in doses required to produce similar effects.

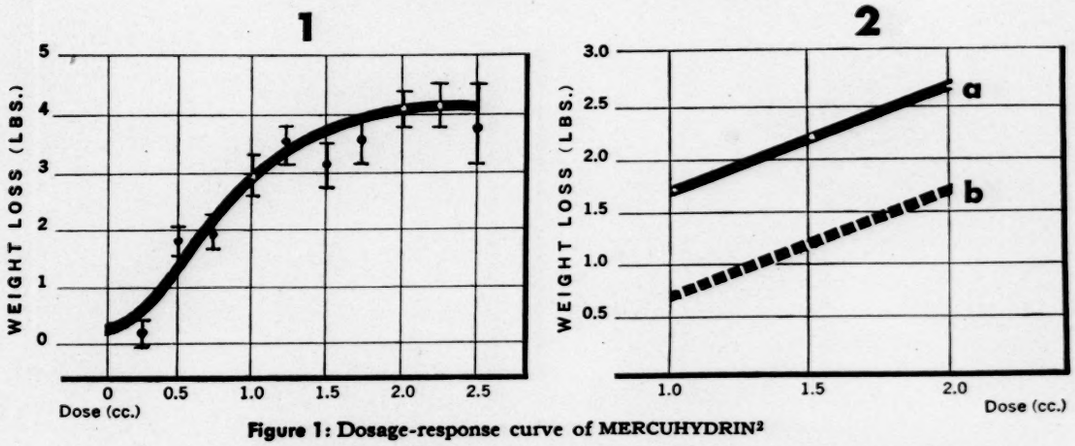


Figure 1: Dosage-response curve of MERCUHYDRIN[2]

Figure 2: Dosage-response curves showing relative diuretic potency of (a) (MERCUHYDRIN) and (b) N-(beta-hydroxymercuri-gamma-methoxypropyl)-2-pyridone-5-carboxylate.[2]

"... to obtain a diuretic response equal to that of meralluride (MERCUHYDRIN) the other mercurial must be given in twice the dosage."[2]

MERCUHYDRIN®
sodium

effective, well tolerated...
intramuscularly, intravenously and subcutaneously

MERCUHYDRIN (Meralluride sodium solution) is available in 1 cc. and 2 cc. ampuls.

bibliography: (1) Clarke, D. A.; Modell, W.; Greiner, T.; Kwit, N. T.; Gluck, J. L.; and Gold, H.: The Dosage-Response Curve for the Comparison of Mercurial Diuretics, Am. J. M. Sc. *220*:156, 1950. (2) Cattell, McK.: Dosage in the Therapy of Cardiovascular Disease, J.A.M.A. *144*:889, 1950.

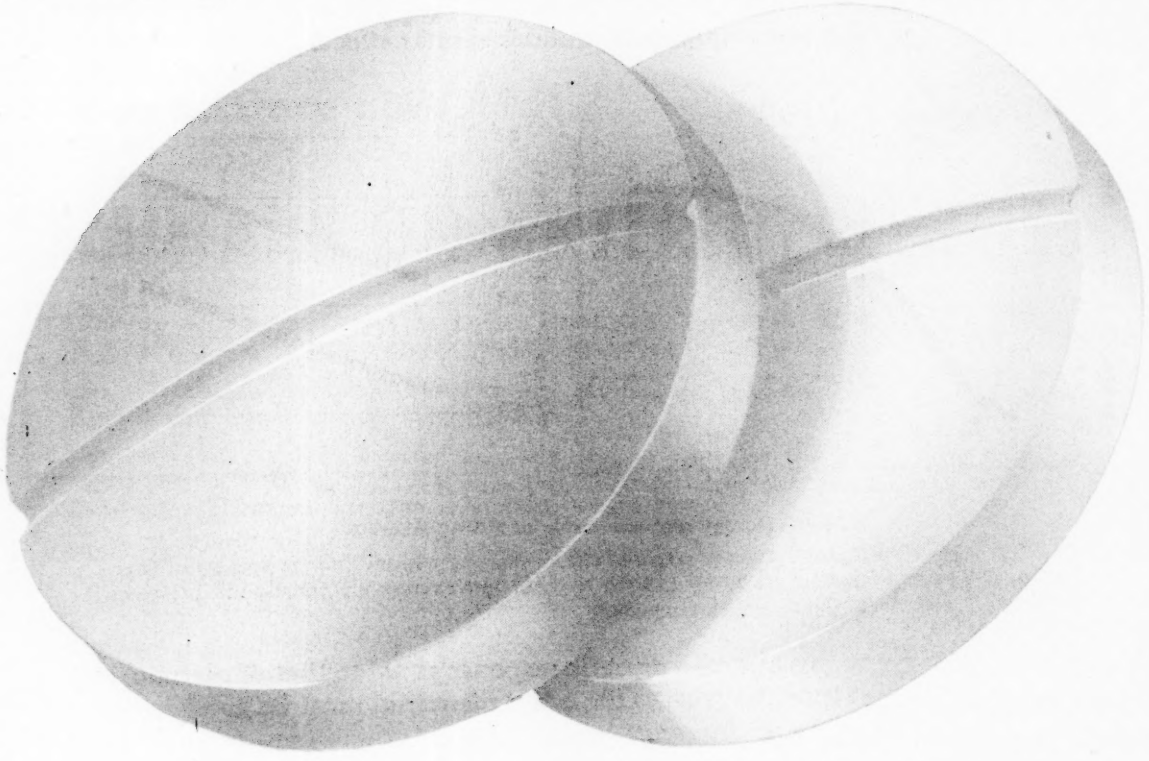
Lakeside Laboratories, INC., MILWAUKEE 1, WISCONSIN

M-16

NOW

2 Banthine Dosage Forms:

THE TRUE ANTICHOLINERGIC IN PEPTIC ULCER IS NOW AVAILABLE AS:



Banthine *and* Banthine with Phenobarbital

"More and more importance is being attached to the role of hypermotility of the stomach in the causation of ulcer pain and less to the influence of simple hyperacidity

"Banthine studies have demonstrated that while it does lower gastric acidity in most patients, its most important function is this same decrease in hypermotility with almost immediate relief of pain."*

DOSAGE FORMS:

Banthine (50 mg.), scored tablets.

Banthine (50 mg.) with Phenobarbital (15 mg.), scored tablets.

*Editorial: Promise in Peptic Ulcer, West. J. Surg. 58:445 (Aug.) 1950.

Banthine®

BROMIDE

BRAND OF METHANTHELINE BROMIDE

SEARLE RESEARCH IN THE SERVICE OF MEDICINE